

| L Number | Hits  | Search Text                                                           | DB                                      | Time stamp       |
|----------|-------|-----------------------------------------------------------------------|-----------------------------------------|------------------|
| 1        | 543   | 536/55.1                                                              | USPAT;<br>US-PPGPUB                     | 2003/09/03 14:13 |
| 2        | 116   | 536/55.1 and chondroitin                                              | USPAT;<br>US-PPGPUB                     | 2003/09/03 14:52 |
| 3        | 15    | (536/55.1 and chondroitin) and recombinant                            | USPAT;<br>US-PPGPUB                     | 2003/09/03 14:15 |
| 4        | 96    | (536/55.1 and chondroitin) and composition                            | USPAT;<br>US-PPGPUB                     | 2003/09/03 14:26 |
| 5        | 65    | ((536/55.1 and chondroitin) and composition)<br>and purified          | USPAT;<br>US-PPGPUB                     | 2003/09/03 14:27 |
| 6        | 26040 | chondroitin polysaccharide                                            | USPAT;<br>US-PPGPUB                     | 2003/09/03 14:52 |
| 7        | 41812 | chondroitin polysaccharide                                            | USPAT;<br>US-PPGPUB;<br>JPO;<br>DERWENT | 2003/09/03 14:52 |
| 8        | 4     | (chondroitin polysaccharide) and chondroitin<br>adj synthase adj gene | USPAT;<br>US-PPGPUB;<br>JPO;            | 2003/09/03 14:53 |
| 9        | 5360  | chondroitin                                                           | DERWENT<br>USPAT;<br>US-PPGPUB;<br>JPO; | 2003/09/03 14:54 |
| 10       | 4     | chondroitin and chondroitin adj synthase<br>adj gene                  | DERWENT<br>USPAT;<br>US-PPGPUB;<br>JPO; | 2003/09/03 14:54 |
| 11       | 4     | chondroitin adj synthase adj gene                                     | DERWENT<br>USPAT;<br>US-PPGPUB;<br>JPO; | 2003/09/03 14:55 |
| 12       | 8     | chondroitin adj synthase                                              | DERWENT<br>USPAT;<br>US-PPGPUB;<br>JPO; | 2003/09/03 14:57 |
| 13       | 1     | chondroitin adj synthetase                                            | DERWENT<br>USPAT;<br>US-PPGPUB;<br>JPO; | 2003/09/03 14:58 |

L20 ANSWER 3 OF 4 CAPLUS COPYRIGHT 2003 ACS  
 ACCESSION NUMBER: 1995:992463 CAPLUS  
 DOCUMENT NUMBER: 124:37797  
 TITLE: Polymers of unsaturated carbohydrate  
       derivatives for use in biomedical articles  
 INVENTOR(S): Bachmann, Frank; Lohmann, Dieter; Chabrecek, Peter  
 PATENT ASSIGNEE(S): Ciba-Geigy A.-G., Switz.  
 SOURCE: Eur. Pat. Appl., 23 pp.  
 CODEN: EPXXDW  
 DOCUMENT TYPE: Patent  
 LANGUAGE: German  
 FAMILY ACC. NUM. COUNT: 1  
 PATENT INFORMATION:

| PATENT NO.                                                                      | KIND | DATE     | APPLICATION NO. | DATE     |
|---------------------------------------------------------------------------------|------|----------|-----------------|----------|
| EP 668294                                                                       | A1   | 19950823 | EP 1995-810079  | 19950207 |
| EP 668294                                                                       | B1   | 19990818 |                 |          |
| R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LI, LU, NL, PT, SE,<br>AT 183517 | E    | 19990915 | AT 1995-810079  | 19950207 |
| AU 9511644                                                                      | A1   | 19950824 | AU 1995-11644   | 19950208 |
| AU 695525                                                                       | B2   | 19980813 |                 |          |
| CA 2142435                                                                      | AA   | 19950816 | CA 1995-2142435 | 19950213 |
| FI 9500623                                                                      | A    | 19950816 | FI 1995-623     | 19950213 |
| JP 07278204                                                                     | A2   | 19951024 | JP 1995-24108   | 19950213 |
| US 5693768                                                                      | A    | 19971202 | US 1995-388006  | 19950213 |
| ZA 9501169                                                                      | A    | 19950815 | ZA 1995-1169    | 19950214 |
| NO 9500548                                                                      | A    | 19950816 | NO 1995-548     | 19950214 |
| CN 1126213                                                                      | A    | 19960710 | CN 1995-101516  | 19950214 |
| US 5856416                                                                      | A    | 19990105 | US 1997-867747  | 19970603 |
| PRIORITY APPLN. INFO.:                                                          |      |          | EP 1994-810084  | 19940215 |
|                                                                                 |      |          | US 1995-388006  | 19950213 |

OTHER SOURCE(S): CASREACT 124:37797; MARPAT 124:37797

GI For diagram(s), see printed CA Issue.

AB Carbohydrate derivs. R<sub>1</sub>(CO<sub>2</sub>A)<sub>m</sub>(O<sub>2</sub>CNHR)<sub>n</sub>(NHCO)pYZ (R<sub>1</sub> = carbohydrate group subject to radical polymn.; A = C<sub>1</sub>-10 alkylene; R = C<sub>1</sub>toreq.20 org. diisocyanate radical; Y = O, NH; Z = mono- or oligosaccharide, cyclodextrin, anhydrosaccharide; m, n, p = 0, 1) can be used in prodn. of polymers useful for contact lenses and other biomedical articles, drug delivery systems, membranes, photoresists, etc. The monomers are obtained by selective reaction of an unprotected carbohydrate with an unsatd. compd., esp. an isocyanate, to form a monosubstituted deriv. Thus, contact lenses of poly(hydroxyethyl méthacrylate) were surface treated with I [reaction product of 4'-(.beta.-hydroxyethoxy)-2-hydroxyprop-2-ylphenone and isophorone diisocyanate], then soaked in an aq. soln. of 6-O-carbamoylmethacryloylethyl-.alpha..alpha.-trehalose (prepn. given), purged to remove O, exposed to an Hg lamp, washed, and dried. The treated lenses showed improved hydrophilicity and water retention.

L24 ANSWER 3 OF 3 CAPLUS COPYRIGHT 2003 ACS

ACCESSION NUMBER: 1975:479501 CAPLUS

DOCUMENT NUMBER: 83:79501

TITLE: Synthesis of oligosaccharides on polymeric supports. V. Selective cleavage by hydrazine of the anomeric acetyl groups of acetylated glycosyl residues

AUTHOR(S): Excoffier, Gerard; Gagnare, Didier; Utile, Jean P.

CORPORATE SOURCE: Cent. Rech. Macromol. Veg., Grenoble, Fr.

SOURCE: Carbohydrate Research (1975), 39(2), 368-73

CODEN: CRBRAT; ISSN: 0008-6215

DOCUMENT TYPE: Journal

LANGUAGE: French

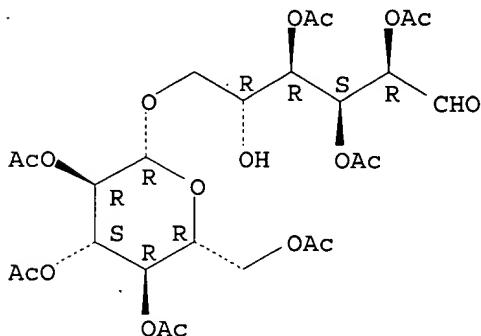
IT 56253-33-9P 56285-96-2P

RL: SPN (Synthetic preparation); PREP (Preparation)  
(prep. of)

RN 56253-33-9 CAPLUS

CN D-Glucose, 6-O-(2,3,4,6-tetra-O-acetyl-.beta.-D-glucopyranosyl)-,  
2,3,4-triacetate (9CI) (CA INDEX NAME)

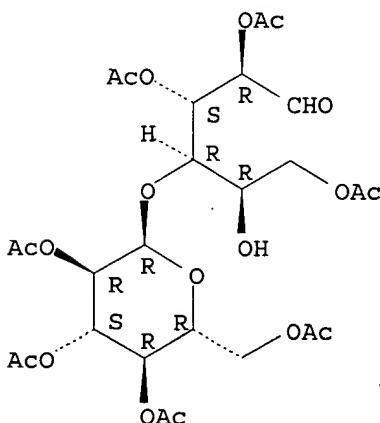
Absolute stereochemistry.



RN 56285-96-2 CAPLUS

CN D-Glucose, 4-O-(2,3,4,6-tetra-O-acetyl-.alpha.-D-glucopyranosyl)-,  
2,3,6-triacetate (9CI) (CA INDEX NAME)

Absolute stereochemistry.



L7 ANSWER 7 OF 7 MEDLINE on STN  
ACCESSION NUMBER: 97306353 MEDLINE  
DOCUMENT NUMBER: 97306353 PubMed ID: 9162078  
TITLE: Biosynthesis of chondroitin sulfate. Purification of glucuronosyl transferase II and use of photoaffinity labeling for characterization of the enzyme as an 80-kDa protein.  
AUTHOR: Sugumaran G; Katsman M; Sunthankar P; Drake R R  
CORPORATE SOURCE: Edith Nourse Rogers Memorial Veterans Hospital, Bedford, Massachusetts 01730, USA.  
CONTRACT NUMBER: AR-41649 (NIAMS)  
SOURCE: JOURNAL OF BIOLOGICAL CHEMISTRY, (1997 May 30) 272 (22) 14399-403.  
Journal code: 2985121R. ISSN: 0021-9258.  
PUB. COUNTRY: United States  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 199706  
ENTRY DATE: Entered STN: 19970716  
Last Updated on STN: 19970716  
Entered Medline: 19970627  
AB A photoaffinity analogue, [ $\beta$ -32P]5-azido-UDP-GlcA, was used to photolabel the enzymes that utilize UDP-GlcA in cartilage microsomes and rat liver microsomes. SDS-polyacrylamide gel electrophoresis analysis of photolabeled cartilage microsomes, which are specialized in chondroitin sulfate synthesis, showed a major radiolabeled band at 80 kDa and other minor radiolabeled bands near 40 and 60 kDa. Rat liver microsomes, which are enriched for enzymes of detoxification by glucuronidation, had a different pattern with multiple major labeled bands near 50-60 and 35 kDa. To determine that the photolabeled 80-kDa protein is the GlcA transferase II, we have purified the enzyme from cartilage microsomes. This membrane-bound enzyme, involved in the transfer of GlcA residues to non-reducing terminal GalNAc residues of the chondroitin polymer, has now been solubilized, stabilized, and then purified greater than 1350-fold by sequential chromatography on Q-Sepharose, heparin-Sepharose, and WGA-agarose. The purified enzyme exhibited a conspicuous silver-stained protein band on SDS-polyacrylamide gel electrophoresis that coincided with the major radiolabeled band of 80 kDa. SDS-polyacrylamide gel analysis of photoaffinity-labeled active fractions from the Q-Sepharose, heparin-Sepharose, and WGA-agarose also indicated only the single radiolabeled band at 80 kDa. Intensity of photolabeling in each of the fractions examined coincided with enzyme activity. The photolabeling of this 80-kDa protein was saturable with the photoprobe and could be inhibited by the addition of UDP-GlcA prior to the addition of the photoprobe. Thus, the photolabeling with [ $\beta$ -32P]5-azido-UDP-GlcA has identified the GlcA transferase II as an 80-kDa protein. The purified enzyme was capable of transferring good amounts of GlcA residues to chondroitin-derived pentasaccharide with negligible transfer to pentasaccharides derived from hyaluronan or heparan.

L17 ANSWER 5 OF 5 MEDLINE on STN  
ACCESSION NUMBER: 85203849 MEDLINE  
DOCUMENT NUMBER: 85203849 PubMed ID: 3922754  
TITLE: Two N-acetylgalactosaminyltransferase are involved in the biosynthesis of chondroitin sulfate.  
AUTHOR: Rohrmann K; Niemann R; Buddecke E  
SOURCE: EUROPEAN JOURNAL OF BIOCHEMISTRY, (1985 May 2) 148 (3)  
463-9.  
Journal code: 0107600. ISSN: 0014-2956.  
PUB. COUNTRY: GERMANY, WEST: Germany, Federal Republic of  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 198507  
ENTRY DATE: Entered STN: 19900320  
Last Updated on STN: 19900320  
Entered Medline: 19850705

AB Two N-acetylgalactosaminyltransferases, designated I and II, have been purified from the microsomal fraction of calf arterial tissue and separated on Bio-Gel A. N-Acetylgalactosaminyltransferase I was purified 450-fold. It requires Mn<sup>2+</sup> for maximal activity and transfers N-acetylgalactosamine residues from UDP-[1-3H]GalNAc in beta-glycosidic configuration to the non-reducing terminus of the acceptor substrates GlcA(beta 1-3)Gal(beta 1-3)Gal, GlcA(beta 1-3)Gal(beta 1-4)Glc and GlcA(beta 1-3)Gal. Even-numbered chondroitin oligosaccharides serve as acceptors for N-acetylgalactosaminyltransferase II, which transfers N-acetylgalactosamine from UDP-[1-3H]GalNAc to the non-reducing glucuronic acid residues of oligosaccharide acceptor substrates. Maximum transfer rates were obtained with a decasaccharide derived from chondroitin. Longer or shorter-chain chondroitin oligosaccharides are less effective acceptor substrates. All reaction products formed by N-acetylgalactosaminyltransferases I and II are substrates of beta-N-acetylhexosaminidase, which splits off the transferred [1-3H]GalNAc completely. In the microsomal fraction N-acetylgalactosaminyltransferase II had a 300-fold higher specific activity than N-acetylgalactosaminyltransferase I. In contrast to enzyme I, enzyme II loses much of its activity during the purification procedure and undergoes rapid thermodenaturation. GlcA-Gal-Gal is a characteristic sequence of the carbohydrate-protein linkage region of proteochondroitin sulfate. The acceptor capacity of this trisaccharide suggests that N-acetylgalactosaminyltransferase I is involved in the synthesis of the carbohydrate-protein linkage region. Since N-acetylgalactosaminyltransferase II is highly specific for chondroitin oligosaccharides, we conclude that it participates in chain elongation during chondroitin sulfate synthesis.

L17 ANSWER 5 OF 5 MEDLINE on STN  
ACCESSION NUMBER: 85203849 MEDLINE  
DOCUMENT NUMBER: 85203849 PubMed ID: 3922754  
TITLE: Two N-acetylgalactosaminyltransferase are involved in the biosynthesis of chondroitin sulfate.  
AUTHOR: Rohrmann K; Niemann R; Buddecke E  
SOURCE: EUROPEAN JOURNAL OF BIOCHEMISTRY, (1985 May 2) 148 (3) 463-9.  
PUB. COUNTRY: GERMANY, WEST: Germany, Federal Republic of  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 198507  
ENTRY DATE: Entered STN: 19900320  
Last Updated on STN: 19900320  
Entered Medline: 19850705

AB Two N-acetylgalactosaminyltransferases, designated I and II, have been purified from the microsomal fraction of calf arterial tissue and separated on Bio-Gel A. N-Acetylgalactosaminyltransferase I was purified 450-fold. It requires Mn<sup>2+</sup> for maximal activity and transfers N-acetylgalactosamine residues from UDP-[1-3H]GalNAc in beta-glycosidic configuration to the non-reducing terminus of the acceptor substrates GlcA(beta 1-3)Gal(beta 1-3)Gal, GlcA(beta 1-3)Gal(beta 1-4)Glc and GlcA(beta 1-3)Gal. Even-numbered chondroitin oligosaccharides serve as acceptors for N-acetylgalactosaminyltransferase II, which transfers N-acetylgalactosamine from UDP-[1-3H]GalNAc to the non-reducing glucuronic acid residues of oligosaccharide acceptor substrates. Maximum transfer rates were obtained with a decasaccharide derived from chondroitin. Longer or shorter-chain chondroitin oligosaccharides are less effective acceptor substrates. All reaction products formed by N-acetylgalactosaminyltransferases I and II are substrates of beta-N-acetylhexosaminidase, which splits off the transferred [1-3H]GalNAc completely. In the microsomal fraction N-acetylgalactosaminyltransferase II had a 300-fold higher specific activity than N-acetylgalactosaminyltransferase I. In contrast to enzyme I, enzyme II loses much of its activity during the purification procedure and undergoes rapid thermodenaturation. GlcA-Gal-Gal is a characteristic sequence of the carbohydrate-protein linkage region of proteochondroitin sulfate. The acceptor capacity of this trisaccharide suggests that N-acetylgalactosaminyltransferase I is involved in the synthesis of the carbohydrate-protein linkage region. Since N-acetylgalactosaminyltransferase II is highly specific for chondroitin oligosaccharides, we conclude that it participates in chain elongation during chondroitin sulfate synthesis.

L17 ANSWER 4 OF 5 MEDLINE on STN  
ACCESSION NUMBER: 1999421651 MEDLINE  
DOCUMENT NUMBER: 99421651 PubMed ID: 10491092  
TITLE: Purification and characterization of fetal bovine serum  
beta-N-acetyl-D-galactosaminyltransferase and  
beta-D-glucuronyltransferase involved in chondroitin  
sulfate biosynthesis.  
AUTHOR: Tsuchida K; Lind T; Kitagawa H; Lindahl U; Sugahara K;  
Lidholt K  
CORPORATE SOURCE: Department of Biochemistry, Kobe Pharmaceutical University,  
Japan.  
SOURCE: EUROPEAN JOURNAL OF BIOCHEMISTRY, (1999 Sep) 264 (2) 461-7.  
Journal code: 0107600. ISSN: 0014-2956.  
PUB. COUNTRY: GERMANY: Germany, Federal Republic of  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 199910  
ENTRY DATE: Entered STN: 19991026  
Last Updated on STN: 19991026  
Entered Medline: 19991014

AB beta-N-Acetylgalactosaminyltransferase II and beta-glucuronyltransferase  
II, involved in chondroitin sulfate biosynthesis, transfer an  
N-acetylgalactosamine (GalNAc) and glucuronic acid (GlcA) residue,  
respectively, through beta-linkages to an acceptor **chondroitin**  
**oligosaccharide** derived from the repeating disaccharide region of  
chondroitin sulfate. They were copurified from fetal bovine serum  
approximately 2500-fold and 850-fold, respectively, by sequential  
chromatographies on Red A-agarose, phenyl-Sepharose, S-Sepharose and wheat  
germ agglutinin-agarose. Identical and inseparable chromatographic  
profiles of both glycosyltransferase activities obtained through the above  
chromatographic steps and gel filtration suggest that the **purified**  
enzyme activities are tightly coupled, which could imply a single enzyme  
with dual transferase activities; beta-N-acetylgalactosaminyltransferase  
and beta-glucuronyltransferase, reminiscent of the heparan sulfate  
polymerase reaction. However, when a polymerization reaction was  
performed in vitro with the **purified** serum enzyme preparation  
under the polymerization conditions recently developed for the  
chondroitin-synthesizing system, derived from human melanoma cells, each  
monosaccharide transfer took place, but no polymerization occurred. These  
results may suggest that the **purified** serum enzyme preparation  
contains both beta-N-acetylgalactosaminyltransferase II and  
beta-glucuronyltransferase II activities on a single polypeptide or on the  
respective polypeptides forming an enzyme complex, but is different from  
that obtained from melanoma cells in that it transfers a single GalNAc or  
GlcA residue but does not polymerize chondroitin.

L6 ANSWER 1 OF 2 CAPLUS COPYRIGHT 2003 ACS on STN  
ACCESSION NUMBER: 2003:435234. CAPLUS  
DOCUMENT NUMBER: 139:18389  
TITLE: Chondroitin synthase gene from *Pasteurella multocida*  
and methods of making and using same  
INVENTOR(S): Deangelis, Paul L.  
PATENT ASSIGNEE(S): USA  
SOURCE: U.S. Pat. Appl. Publ., 38 pp., Cont.-in-part of U.S.  
Ser. No. 437,277.  
CODEN: USXXCO  
DOCUMENT TYPE: Patent  
LANGUAGE: English  
FAMILY ACC. NUM. COUNT: 12  
PATENT INFORMATION:

| PATENT NO.             | KIND | DATE     | APPLICATION NO. | DATE        |
|------------------------|------|----------|-----------------|-------------|
| US 2003104601          | A1   | 20030605 | US 2001-842484  | 20010425    |
| US 6444447             | B1   | 20020903 | US 1999-437277  | 19991110    |
| US 2003113845          | A1   | 20030619 | US 2002-217613  | 20020812    |
| PRIORITY APPLN. INFO.: |      |          |                 |             |
|                        |      |          | US 1999-283402  | B2 19990401 |
|                        |      |          | US 1999-437277  | A2 19991110 |
|                        |      |          | US 2000-199538P | P 20000425  |
|                        |      |          | US 1998-178851  | B2 19981026 |
|                        |      |          | US 1998-107929P | P 19981111  |

AB The present invention relates to a chondroitin synthase gene and methods of making and using same. In more particular, but not by way of limitation, the present invention relates to a chondroitin synthase gene from *Pasteurella multocida* and methods of isolating and using same. Addnl., the present invention relates to the use of unsulfated chondroitin and its prepn., as well as conversion into modified versions such as dermatan sulfate and chondroitin sulfate polymers.

L6 ANSWER 2 OF 2 CAPLUS COPYRIGHT 2003 ACS on STN DUPLICATE 1  
ACCESSION NUMBER: 2002:888388 CAPLUS  
DOCUMENT NUMBER: 138:382456  
TITLE: Biosynthesis of Chondroitin/Dermatan Sulfate  
AUTHOR(S): Silbert, Jeremiah E.; Sugumaran, Geetha  
CORPORATE SOURCE: Dep. Veterans Affairs Med. Cent., Bedford, MA, 01730,  
USA  
SOURCE: IUBMB Life (2002), 54(4), 177-186  
CODEN: IULIF8; ISSN: 1521-6543  
PUBLISHER: Taylor & Francis Inc.  
DOCUMENT TYPE: Journal; General Review  
LANGUAGE: English

AB A review. Chondroitin sulfate and dermatan sulfate are synthesized as galactosaminoglycan polymers contg. N-acetylgalactosamine alternating with glucuronic acid. The sugar residues are sulfated to varying degrees and positions depending upon the tissue sources and varying conditions of formation. Epimerization of any of the glucuronic acid residues to iduronic acid at the polymer level constitutes the formation of dermatan sulfate. Chondroitin/dermatan glycosaminoglycans are covalently attached by a common tetrasaccharide sequence to the serine residues of core proteins while they are adherent to the inner surface of endoplasmic reticulum/Golgi vesicles. Addn. of the first sugar residue, xylose, to core proteins begins in the endoplasmic reticulum, followed by the addn. of two galactose residues by two distinct glycosyl transferases in the early cis/medial regions of the Golgi. The linkage tetrasaccharide is completed in the medial/trans Golgi by the addn. of the first glucuronic acid residue, followed by transfer of N-acetylgalactosamine to initiate the formation of a galactosaminoglycan rather than a glucosaminoglycan. This specific N-acetylgalactosaminyl transferase is different from the chondroitin synthase involved in generation of the

repeating disaccharide units to form the **chondroitin polymer**. Sulfation of the **chondroitin polymer** by specific sulfotransferases occurs as the polymer is being formed. All the enzymes in the pathway for synthesis have been cloned, with the exception of the glucuronyl to iduronyl epimerase involved in the formation of dermatan residues.

REFERENCE COUNT: 82 THERE ARE 82 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

> d his

(FILE 'HOME' ENTERED AT 15:11:28 ON 03 SEP 2003)

FILE 'CAPLUS, MEDLINE' ENTERED AT 15:11:36 ON 03 SEP 2003

L1           0 S CHRONDROITIN POLYMER  
L2           10 S CHONDROITIN POLYMER  
L3           0 S L2 AND COMPOSITION  
L4           7 S L2 AND CHONDROITIN SYNTHASE  
L5           3 S L2 AND SULFATED  
L6           2 DUP REM L5 (1 DUPLICATE REMOVED)

L7 ANSWER 7 OF 7 MEDLINE on STN  
ACCESSION NUMBER: 97306353 MEDLINE  
DOCUMENT NUMBER: 97306353 PubMed ID: 9162078  
TITLE: Biosynthesis of chondroitin sulfate. Purification of glucuronosyl transferase II and use of photoaffinity labeling for characterization of the enzyme as an 80-kDa protein.  
AUTHOR: Sugumaran G; Katsman M; Sunthankar P; Drake R R  
CORPORATE SOURCE: Edith Nourse Rogers Memorial Veterans Hospital, Bedford, Massachusetts 01730, USA.  
CONTRACT NUMBER: AR-41649 (NIAMS)  
SOURCE: JOURNAL OF BIOLOGICAL CHEMISTRY, (1997 May 30) 272 (22) 14399-403.  
Journal code: 2985121R. ISSN: 0021-9258.  
PUB. COUNTRY: United States  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 199706  
ENTRY DATE: Entered STN: 19970716  
Last Updated on STN: 19970716  
Entered Medline: 19970627

AB A photoaffinity analogue, [ $\beta$ -32P]5-azido-UDP-GlcA, was used to photolabel the enzymes that utilize UDP-GlcA in cartilage microsomes and rat liver microsomes. SDS-polyacrylamide gel electrophoresis analysis of photolabeled cartilage microsomes, which are specialized in chondroitin sulfate synthesis, showed a major radiolabeled band at 80 kDa and other minor radiolabeled bands near 40 and 60 kDa. Rat liver microsomes, which are enriched for enzymes of detoxification by glucuronidation, had a different pattern with multiple major labeled bands near 50-60 and 35 kDa. To determine that the photolabeled 80-kDa protein is the GlcA transferase II, we have purified the enzyme from cartilage microsomes. This membrane-bound enzyme, involved in the transfer of GlcA residues to non-reducing terminal GalNAc residues of the chondroitin polymer, has now been solubilized, stabilized, and then purified greater than 1350-fold by sequential chromatography on Q-Sepharose, heparin-Sepharose, and WGA-agarose. The purified enzyme exhibited a conspicuous silver-stained protein band on SDS-polyacrylamide gel electrophoresis that coincided with the major radiolabeled band of 80 kDa. SDS-polyacrylamide gel analysis of photoaffinity-labeled active fractions from the Q-Sepharose, heparin-Sepharose, and WGA-agarose also indicated only the single radiolabeled band at 80 kDa. Intensity of photolabeling in each of the fractions examined coincided with enzyme activity. The photolabeling of this 80-kDa protein was saturable with the photoprobe and could be inhibited by the addition of UDP-GlcA prior to the addition of the photoprobe. Thus, the photolabeling with [ $\beta$ -32P]5-azido-UDP-GlcA has identified the GlcA transferase II as an 80-kDa protein. The purified enzyme was capable of transferring good amounts of GlcA residues to chondroitin-derived pentasaccharide with negligible transfer to pentasaccharides derived from hyaluronan or heparan.

L7 ANSWER 1 OF 7 CAPLUS COPYRIGHT 2003 ACS on STN  
 ACCESSION NUMBER: 2002:669614 CAPLUS  
 DOCUMENT NUMBER: 137:215875  
 TITLE: Polymer grafting with polysaccharide synthases for  
       coating biomaterial surfaces  
 INVENTOR(S): DeAngelis, Paul L.  
 PATENT ASSIGNEE(S): The Board of Regents of the University of Oklahoma,  
       USA  
 SOURCE: U.S., 37 pp., Cont.-in-part of U.S. Provisional Ser.  
       No. 283,402.  
 CODEN: USXXAM  
 DOCUMENT TYPE: Patent  
 LANGUAGE: English  
 FAMILY ACC. NUM. COUNT: 12  
 PATENT INFORMATION:

| PATENT NO.                                                                                                                                                                                                                                                                                | KIND | DATE     | APPLICATION NO. | DATE     |
|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|------|----------|-----------------|----------|
| US 6444447                                                                                                                                                                                                                                                                                | B1   | 20020903 | US 1999-437277  | 19991110 |
| WO 2001080810                                                                                                                                                                                                                                                                             | A2   | 20011101 | WO 2001-US13395 | 20010425 |
| WO 2001080810                                                                                                                                                                                                                                                                             | A3   | 20020516 |                 |          |
| W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN,<br>CO, CR, CU, CZ, DE, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM,<br>HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS,<br>LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO,<br>RU, SD, SE |      |          |                 |          |
| RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY,<br>DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF,<br>BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG                                                                                          |      |          |                 |          |
| US 2003104601                                                                                                                                                                                                                                                                             | A1   | 20030605 | US 2001-842484  | 20010425 |
| US 2003077763                                                                                                                                                                                                                                                                             | A1   | 20030424 | US 2002-184485  | 20020627 |
| US 2003113845                                                                                                                                                                                                                                                                             | A1   | 20030619 | US 2002-217613  | 20020812 |
| PRIORITY APPLN. INFO.:                                                                                                                                                                                                                                                                    |      |          |                 |          |
| US 1998-107929P P 19981111                                                                                                                                                                                                                                                                |      |          |                 |          |
| US 1999-283402 A2 19990401                                                                                                                                                                                                                                                                |      |          |                 |          |
| US 1998-178851 B2 19981026                                                                                                                                                                                                                                                                |      |          |                 |          |
| US 1999-437277 A2 19991110                                                                                                                                                                                                                                                                |      |          |                 |          |
| US 2000-199538P P 20000425                                                                                                                                                                                                                                                                |      |          |                 |          |

AB The present invention relates to methodol. for polymer grafting by a  
 polysaccharide synthase and, more particularly, polymer grafting using the  
 hyaluronate synthase from Pasteurella multocida. The present invention  
 also relates to coatings for biomaterials wherein the coatings provide  
 protective properties to the biomaterial and/or act as a bioadhesive.  
 Such coatings could be applied to elec. devices, sensors, catheters and  
 any device which may be contemplated for use within a mammal. The present  
 invention further relates to drug delivery matrixes which are  
 biocompatible and may comprise combinations of a biomaterial or a  
 bioadhesive and a medicament or a medicament-contg. liposome. The  
 biomaterial and/or bioadhesive is a hyaluronic acid polymer produced by a  
 hyaluronate synthase from Pasteurella multocida. The present invention  
 also relates to the creation of chimeric mols. contg. hyaluronic acid or  
 hyaluronic acid-like chains attached to various compds. and esp.  
 carbohydrates or hydroxyl contg. substances. The present invention also  
 relates to a chondroitin synthase from Pasteurella multocida which is  
 capable of producing polysaccharide polymers on an acceptor or primer mol.

REFERENCE COUNT: 30 THERE ARE 30 CITED REFERENCES AVAILABLE FOR THIS  
 RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 2 OF 7 CAPLUS COPYRIGHT 2003 ACS on STN  
 ACCESSION NUMBER: 2002:478986 CAPLUS  
 DOCUMENT NUMBER: 137:243778  
 TITLE: Molecular cloning and characterization of chondroitin  
       polymerase from Escherichia coli strain K4  
 AUTHOR(S): Ninomiya, Toshio; Sugiura, Nobuo; Tawada, Akira;

CORPORATE SOURCE: Sugimoto, Kazunori; Watanabe, Hideto; Kimata, Koji  
 Institute for Molecular Science of Medicine, Aichi  
 Medical University, Nagakute, 480-1195, Japan  
 SOURCE: Journal of Biological Chemistry (2002), 277(24),  
 21567-21575  
 CODEN: JBCHA3; ISSN: 0021-9258  
 PUBLISHER: American Society for Biochemistry and Molecular  
 Biology  
 DOCUMENT TYPE: Journal  
 LANGUAGE: English  
 AB Escherichia coli strain K4 produces the K4 antigen, a capsule polysaccharide consisting of a chondroitin backbone (GlcUA .beta.(1-3)-GalNAc .beta.(1-4))n to which .beta.-fructose is linked at position C-3 of the GlcUA residue. The authors molecularly cloned region 2 of the K4 capsular gene cluster essential for biosynthesis of the polysaccharide, and the authors further identified a gene encoding a bifunctional glycosyltransferase that polymerizes the chondroitin backbone. The enzyme, contg. two conserved glycosyltransferase sites, showed 59 and 61% identity at the amino acid level to class 2 hyaluronan synthase and chondroitin synthase from Pasteurella multocida, resp. The sol. enzyme expressed in a bacterial expression system transferred GalNAc and GlcUA residues alternately, and polymd. the chondroitin chain up to a mol. mass of 20 kDa when chondroitin sulfate hexasaccharide was used as an acceptor. The enzyme exhibited apparent Km values for UDP-GlcUA and UDP-GalNAc of 3.44 and 31.6 .mu.M, resp., and absolutely required acceptors of chondroitin sulfate polymers and oligosaccharides at least longer than a tetrasaccharide. In addn., chondroitin polymers and oligosaccharides served as acceptors for chondroitin polymn., but dermatan sulfate and heparin did not. These results may lead to elucidation of the mechanism for chondroitin chain synthesis in both microorganisms and mammals.  
 REFERENCE COUNT: 48 THERE ARE 48 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 3 OF 7 CAPLUS COPYRIGHT 2003 ACS on STN  
 ACCESSION NUMBER: 2000:335277 CAPLUS  
 DOCUMENT NUMBER: 133:9149  
 TITLE: Polymer grafting with polysaccharide synthases for coating biomaterial surfaces  
 INVENTOR(S): Deangelis, Paul L.  
 PATENT ASSIGNEE(S): The Board of Regents of the University of Oklahoma,  
 USA  
 SOURCE: PCT Int. Appl., 86 pp.  
 CODEN: PIIXD2  
 DOCUMENT TYPE: Patent  
 LANGUAGE: English  
 FAMILY ACC. NUM. COUNT: 12  
 PATENT INFORMATION:

| PATENT NO.    | KIND                                                                                                                                                                                                                                                                                                                                                       | DATE     | APPLICATION NO. | DATE     |
|---------------|------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|----------|-----------------|----------|
| WO 2000027437 | A2                                                                                                                                                                                                                                                                                                                                                         | 20000518 | WO 1999-US26501 | 19991110 |
| WO 2000027437 | A3                                                                                                                                                                                                                                                                                                                                                         | 20000720 |                 |          |
| W:            | AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CR, CU, CZ, DE, DK, DM, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM |          |                 |          |
| RW:           | GH, GM, KE, LS, MW, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG                                                                                                                                                                             |          |                 |          |
| AU 2000016139 | A5                                                                                                                                                                                                                                                                                                                                                         | 20000529 | AU 2000-16139   | 19991110 |

|                                                                                              |    |          |                 |             |
|----------------------------------------------------------------------------------------------|----|----------|-----------------|-------------|
| EP 1129209                                                                                   | A2 | 20010905 | EP 1999-958858  | 19991110    |
| R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT,<br>IE, SI, LT, LV, FI, RO |    |          |                 |             |
| JP 2002529064                                                                                | T2 | 20020910 | JP 2000-580666  | 19991110    |
| US 2003113845                                                                                | A1 | 20030619 | US 2002-217613  | 20020812    |
| PRIORITY APPLN. INFO.:                                                                       |    |          | US 1998-107929P | P 19981111  |
|                                                                                              |    |          | US 1999-283402  | A 19990401  |
|                                                                                              |    |          | US 1998-178851  | B2 19981026 |
|                                                                                              |    |          | WO 1999-US26501 | W 19991110  |

AB The present invention relates to methodol. for polymer grafting by a polysaccharide synthase and, more particularly, polymer grafting using the hyaluronate synthase from *Pasteurella multocida*. The present invention also relates to coatings for biomaterials wherein the coatings provide protective properties to the biomaterial and/or act as a bioadhesive. Such coatings could be applied to elec. devices, sensors, catheters and any device which may be contemplated for use within a mammal. The present invention further relates to drug delivery matrixes which are biocompatible and may comprise combinations of a biomaterial or a bioadhesive and a medicament or a medicament-contg. liposome. The biomaterial and/or bioadhesive is a hyaluronic acid polymer produced by a hyaluronate synthase from *Pasteurella multocida*. The present invention also relates to the creation of chimeric mols. contg. hyaluronic acid or hyaluronic acid-like chains attached to various compds. and esp. carbohydrates or hydroxyl contg. substances. The present invention also relates to a chondroitin synthase from *Pasteurella multocida* which is capable of producing polysaccharide polymers on an acceptor or primer mol.

L7 ANSWER 4 OF 7 CAPLUS COPYRIGHT 2003 ACS on STN

ACCESSION NUMBER: 1997:358015 CAPLUS

DOCUMENT NUMBER: 127:92009

TITLE: Biosynthesis of chondroitin sulfate. Purification of glucuronosyl transferase II and use of photoaffinity labeling for characterization of the enzyme as an 80-kDa protein

AUTHOR(S): Sugumaran, Geetha; Katsman, Maya; Sunthankar, Prassana; Drake, Richard R.

CORPORATE SOURCE: Edith Nourse Rogers Memorial Veterans Hospital, Bedford, MA, 01730, USA

SOURCE: Journal of Biological Chemistry (1997), 272(22), 14399-14403

CODEN: JBCHA3; ISSN: 0021-9258

PUBLISHER: American Society for Biochemistry and Molecular Biology

DOCUMENT TYPE: Journal

LANGUAGE: English

AB A photoaffinity analog, [ $\beta$ -32P]5-azido-UDP-GlcA, was used to photolabel the enzymes that utilize UDP-GlcA in cartilage microsomes and rat liver microsomes. SDS-PAGE anal. of photolabeled cartilage microsomes, which are specialized in chondroitin sulfate synthesis, showed a major radiolabeled band at 80 kDa and other minor radiolabeled bands near 40 and 60 kDa. Rat liver microsomes, which are enriched for enzymes of detoxification by glucuronidation, had a different pattern with multiple major labeled bands near 50-60 and 35 kDa. To det. that the photolabeled 80-kDa protein is the GlcA transferase II, we have purified the enzyme from cartilage microsomes. This membrane-bound enzyme, involved in the transfer of GlcA residues to non-reducing terminal Gal-NAc residues of the chondroitin polymer, has now been solubilized, stabilized, and then purified greater than 1350-fold by sequential chromatog. on Q-Sepharose, heparin-Sepharose, and WGA-agarose. The purified enzyme exhibited a conspicuous silver-stained protein band on SDS-PAGE that coincided with the major radiolabeled band of 80 kDa. SDS-polyacrylamide gel anal. of photoaffinity-labeled active fractions from the Q-Sepharose, heparin-Sepharose, and WGA-agarose also indicated only the single radiolabeled band at 80 kDa. Intensity of photolabeling

in each of the fractions examined coincided with enzyme activity. The photolabeling of this 80-kDa protein was saturable with the photoprobe and could be inhibited by the addn. of UCP-GlcA prior to the addn. of the photoprobe. Thus, the photolabeling with [ $\beta$ -32P]5-azido-UDP-GlcA has identified the GlcA transferase II as an 80-kDa protein. The purified enzyme was capable of transferring good amounts of GlcA residues to chondroitin-derived pentasaccharide with negligible transfer to pentasaccharides derived from hyaluronan or heparan.

L7 ANSWER 5 OF 7 CAPLUS COPYRIGHT 2003 ACS on STN

ACCESSION NUMBER: 1968:492940 CAPLUS  
DOCUMENT NUMBER: 69:92940  
TITLE: Protein-chondroitine-keratan sulfate hybrid complex  
AUTHOR(S): Bychkov, S. M.; Kharlamova, V. N.  
CORPORATE SOURCE: Nauch.-Issled. Lab. Min. Zdravookhr., Moscow, USSR  
SOURCE: Biokhimiya (Moscow) (1968), 33(4), 840-6  
CODEN: BIOHAO; ISSN: 0320-9725  
DOCUMENT TYPE: Journal  
LANGUAGE: Russian

AB A complex biopolymer formed by hybridization of protein-chondroitin-4-sulfate with keratan sulfate (I) was found in the cartilage from bovine tracheal rings. Proteolytic digestion with papain cleaved off polypeptide-chondroitin-4-sulfate (II). Complete loss of all I from the biopolymer together with >90% loss of its protein component did not affect the ability of II to form a complex with hexaminocobalt-(III)-chloride, Rivanol, lysozyme, or acid-sol. collagen at certain pH value and ionic strength. Rivanol was not suitable for the fractionation of glucosaminoglycans since it formed insol. complexes at pH 2 both with them and with the original complex polymer.

L7 ANSWER 6 OF 7 MEDLINE on STN

ACCESSION NUMBER: 2002330828 MEDLINE  
DOCUMENT NUMBER: 22050655 PubMed ID: 11943778  
TITLE: Molecular cloning and characterization of chondroitin polymerase from Escherichia coli strain K4.  
AUTHOR: Ninomiya Toshio; Sugiura Nobuo; Tawada Akira; Sugimoto Kazunori; Watanabe Hideto; Kimata Koji  
CORPORATE SOURCE: Institute for Molecular Science of Medicine, Aichi Medical University, Yazako, Nagakute, Aichi 480-1195, Japan.  
SOURCE: JOURNAL OF BIOLOGICAL CHEMISTRY, (2002 Jun 14) 277 (24) 21567-75.

PUB. COUNTRY: United States  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
OTHER SOURCE: GENBANK-AB079602; GENBANK-X77617  
ENTRY MONTH: 200207  
ENTRY DATE: Entered STN: 20020621  
Last Updated on STN: 20030105  
Entered Medline: 20020725

AB Escherichia coli strain K4 produces the K4 antigen, a capsule polysaccharide consisting of a chondroitin backbone (GlcUA  $\beta$ (1-3)-GalNAc  $\beta$ (1-4))(n) to which beta-fructose is linked at position C-3 of the GlcUA residue. We molecularly cloned region 2 of the K4 capsular gene cluster essential for biosynthesis of the polysaccharide, and we further identified a gene encoding a bifunctional glycosyltransferase that polymerizes the chondroitin backbone. The enzyme, containing two conserved glycosyltransferase sites, showed 59 and 61% identity at the amino acid level to class 2 hyaluronan synthase and chondroitin synthase from Pasteurella multocida, respectively. The soluble enzyme expressed in a bacterial expression system transferred GalNAc and GlcUA residues alternately, and polymerized the chondroitin chain up to a molecular mass of 20 kDa when chondroitin sulfate

hexasaccharide was used as an acceptor. The enzyme exhibited apparent K(m) values for UDP-GlcUA and UDP-GalNAc of 3.44 and 31.6 microm, respectively, and absolutely required acceptors of chondroitin sulfate polymers and oligosaccharides at least longer than a tetrasaccharide. In addition, **chondroitin polymers** and oligosaccharides and hyaluronan polymers and oligosaccharides served as acceptors for chondroitin polymerization, but dermatan sulfate and heparin did not. These results may lead to elucidation of the mechanism for chondroitin chain synthesis in both microorganisms and mammals.

L7 ANSWER 7 OF 7 MEDLINE on STN  
ACCESSION NUMBER: 97306353 MEDLINE  
DOCUMENT NUMBER: 97306353 PubMed ID: 9162078  
TITLE: Biosynthesis of chondroitin sulfate. Purification of glucuronosyl transferase II and use of photoaffinity labeling for characterization of the enzyme as an 80-kDa protein.  
AUTHOR: Sugumaran G; Katsman M; Sunthankar P; Drake R R  
CORPORATE SOURCE: Edith Nourse Rogers Memorial Veterans Hospital, Bedford, Massachusetts 01730, USA.  
CONTRACT NUMBER: AR-41649 (NIAMS)  
SOURCE: JOURNAL OF BIOLOGICAL CHEMISTRY, (1997 May 30) 272 (22) 14399-403.  
Journal code: 2985121R. ISSN: 0021-9258.  
PUB. COUNTRY: United States  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 199706  
ENTRY DATE: Entered STN: 19970716  
Last Updated on STN: 19970716  
Entered Medline: 19970627  
AB A photoaffinity analogue, [ $\beta$ -32P]5-azido-UDP-GlcA, was used to photolabel the enzymes that utilize UDP-GlcA in cartilage microsomes and rat liver microsomes. SDS-polyacrylamide gel electrophoresis analysis of photolabeled cartilage microsomes, which are specialized in chondroitin sulfate synthesis, showed a major radiolabeled band at 80 kDa and other minor radiolabeled bands near 40 and 60 kDa. Rat liver microsomes, which are enriched for enzymes of detoxification by glucuronidation, had a different pattern with multiple major labeled bands near 50-60 and 35 kDa. To determine that the photolabeled 80-kDa protein is the GlcA transferase II, we have purified the enzyme from cartilage microsomes. This membrane-bound enzyme, involved in the transfer of GlcA residues to non-reducing terminal GalNAc residues of the **chondroitin polymer**, has now been solubilized, stabilized, and then purified greater than 1350-fold by sequential chromatography on Q-Sepharose, heparin-Sepharose, and WGA-agarose. The purified enzyme exhibited a conspicuous silver-stained protein band on SDS-polyacrylamide gel electrophoresis that coincided with the major radiolabeled band of 80 kDa. SDS-polyacrylamide gel analysis of photoaffinity-labeled active fractions from the Q-Sepharose, heparin-Sepharose, and WGA-agarose also indicated only the single radiolabeled band at 80 kDa. Intensity of photolabeling in each of the fractions examined coincided with enzyme activity. The photolabeling of this 80-kDa protein was saturable with the photoprobe and could be inhibited by the addition of UDP-GlcA prior to the addition of the photoprobe. Thus, the photolabeling with [ $\beta$ -32P]5-azido-UDP-GlcA has identified the GlcA transferase II as an 80-kDa protein. The purified enzyme was capable of transferring good amounts of GlcA residues to chondroitin-derived pentasaccharide with negligible transfer to pentasaccharides derived from hyaluronan or heparan.

L7 ANSWER 7 OF 7 MEDLINE on STN  
ACCESSION NUMBER: 97306353 MEDLINE  
DOCUMENT NUMBER: 97306353 PubMed ID: 9162078  
TITLE: Biosynthesis of chondroitin sulfate. Purification of glucuronosyl transferase II and use of photoaffinity labeling for characterization of the enzyme as an 80-kDa protein.  
AUTHOR: Sugumaran G; Katsman M; Sunthankar P; Drake R R  
CORPORATE SOURCE: Edith Nourse Rogers Memorial Veterans Hospital, Bedford, Massachusetts 01730, USA.  
CONTRACT NUMBER: AR-41649 (NIAMS)  
SOURCE: JOURNAL OF BIOLOGICAL CHEMISTRY, (1997 May 30) 272 (22) 14399-403.  
Journal code: 2985121R. ISSN: 0021-9258.  
PUB. COUNTRY: United States  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 199706  
ENTRY DATE: Entered STN: 19970716  
Last Updated on STN: 19970716  
Entered Medline: 19970627

AB A photoaffinity analogue, [ $\beta$ -32P]5-azido-UDP-GlcA, was used to photolabel the enzymes that utilize UDP-GlcA in cartilage microsomes and rat liver microsomes. SDS-polyacrylamide gel electrophoresis analysis of photolabeled cartilage microsomes, which are specialized in chondroitin sulfate synthesis, showed a major radiolabeled band at 80 kDa and other minor radiolabeled bands near 40 and 60 kDa. Rat liver microsomes, which are enriched for enzymes of detoxification by glucuronidation, had a different pattern with multiple major labeled bands near 50-60 and 35 kDa. To determine that the photolabeled 80-kDa protein is the GlcA transferase II, we have purified the enzyme from cartilage microsomes. This membrane-bound enzyme, involved in the transfer of GlcA residues to non-reducing terminal GalNAc residues of the chondroitin polymer, has now been solubilized, stabilized, and then purified greater than 1350-fold by sequential chromatography on Q-Sepharose, heparin-Sepharose, and WGA-agarose. The purified enzyme exhibited a conspicuous silver-stained protein band on SDS-polyacrylamide gel electrophoresis that coincided with the major radiolabeled band of 80 kDa. SDS-polyacrylamide gel analysis of photoaffinity-labeled active fractions from the Q-Sepharose, heparin-Sepharose, and WGA-agarose also indicated only the single radiolabeled band at 80 kDa. Intensity of photolabeling in each of the fractions examined coincided with enzyme activity. The photolabeling of this 80-kDa protein was saturable with the photoprobe and could be inhibited by the addition of UDP-GlcA prior to the addition of the photoprobe. Thus, the photolabeling with [ $\beta$ -32P]5-azido-UDP-GlcA has identified the GlcA transferase II as an 80-kDa protein. The purified enzyme was capable of transferring good amounts of GlcA residues to chondroitin-derived pentasaccharide with negligible transfer to pentasaccharides derived from hyaluronan or heparan.

=> d his

(FILE 'HOME' ENTERED AT 15:11:28 ON 03 SEP 2003)

FILE 'CPLUS, MEDLINE' ENTERED AT 15:11:36 ON 03 SEP 2003

L1           0 S CHRONDROITIN POLYMER  
L2           10 S CHONDROITIN POLYMER  
L3           0 S L2 AND COMPOSITION  
L4           7 S L2 AND CHONDROITIN SYNTHASE  
L5           3 S L2 AND SULFATED  
L6           2 DUP REM L5 (1 DUPLICATE REMOVED)  
L7           7 S L2 NOT L5

L9 ANSWER 1 OF 6 CAPLUS COPYRIGHT 2003 ACS on STN  
 ACCESSION NUMBER: 2003:610628 CAPLUS  
 TITLE: Novel human N-acetylgalactosamine transferase GalNAc-T  
       that transfers N-acetylgalactosamine to glucuronic  
       acid of glycosaminoglycan  
 INVENTOR(S): Narimatsu, Hisashi; Gotoh, Masanori  
 PATENT ASSIGNEE(S): National Institute of Advanced Industrial Science and  
       Technology, Japan; Amersham Biosciences K.K.;  
       Fujirebio Inc.  
 SOURCE: PCT Int. Appl., 61 pp.  
 CODEN: PIXXD2  
 DOCUMENT TYPE: Patent  
 LANGUAGE: Japanese  
 FAMILY ACC. NUM. COUNT: 1  
 PATENT INFORMATION:

| PATENT NO.                                                                                                                                                                                                                                                                                                                                                                                                                  | KIND | DATE     | APPLICATION NO. | DATE       |
|-----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|------|----------|-----------------|------------|
| WO 2003064645                                                                                                                                                                                                                                                                                                                                                                                                               | A1   | 20030807 | WO 2003-JP1023  | 20030131   |
| W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN,<br>CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH,<br>GM, HR, HU, ID, IL, IN, IS, KE, KG, KP, KR, KZ, LC, LK, LR, LS,<br>LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL,<br>PT, RO, RU, SC, SD, SE, SG, SK, SL, TJ, TM, TN, TR, TT, TZ, UA,<br>UG, US, UZ, VC, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU,<br>TJ, TM |      |          |                 |            |
| RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, BG,<br>CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC,<br>NL, PT, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW,<br>ML, MR, NE, SN, TD, TG                                                                                                                                                                                         |      |          |                 |            |
| PRIORITY APPLN. INFO.:                                                                                                                                                                                                                                                                                                                                                                                                      |      |          | JP 2002-24202   | A 20020131 |
|                                                                                                                                                                                                                                                                                                                                                                                                                             |      |          | JP 2002-299309  | A 20021011 |

AB A novel human enzyme transferring N-acetylgalactosamine (GalNAc) to  
   glucuronic acid of glycosaminoglycan, encoding cDNA, recombinant  
   expression, are disclosed. Probes, primers, and microarrays and reagent  
   kits contg. them, for detecting the gene, are claimed. The authors found  
   a novel glycosyltransferase gene having a hypothetical  
   .bet.a.1,4-galactosyltransferase motif by a BLAST search and cloned its  
   full-length open reading frame using the 5'-rapid amplification of cDNA  
   ends method. The truncated form was expressed in insect cells as a sol.  
   enzyme. It transferred N-acetylgalactosamine, not galactose, to  
   para-nitrophenyl-.beta.-glucuronic acid. The N-acetylgalactosamine-  
   glucuronic acid linkage has been identified only in chondroitin sulfate;  
   therefore, the authors examd. its chondroitin elongation and initiation  
   activities. N-Acetylgalactosaminyltransferase activity was obsd. toward  
   chondroitin poly- and oligosaccharides, chondroitin sulfate  
   oligosaccharides, and linkage tetrasaccharide (GlcA-Gal-Gal-Xyl-O-  
   methoxyphenyl), and the chondroitin polysaccharide and  
   linkage tetrasaccharide were better acceptor substrates than the others.  
   These results suggest that this enzyme has N-acetylgalactosaminyltransfера<sup>se</sup>  
   activity in both the elongation and initiation of chondroitin sulfate  
   synthesis.

REFERENCE COUNT: 4 THERE ARE 4 CITED REFERENCES AVAILABLE FOR THIS  
                   RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L9 ANSWER 2 OF 6 CAPLUS COPYRIGHT 2003 ACS on STN  
 ACCESSION NUMBER: 2003:22694 CAPLUS  
 DOCUMENT NUMBER: 138:83382  
 TITLE: Polysaccharides with Helicobacter pylori receptor  
       activity for treatment of gastric diseases  
 INVENTOR(S): Natunen, Jari; Miller-Podraza, Halina; Teneberg,  
       Susann; Angstroem, Jonas; Karlsson, Karl-Anders  
 PATENT ASSIGNEE(S): Carbion Oy, Finland

SOURCE: PCT Int. Appl., 72 pp.  
 CODEN: PIXXD2  
 DOCUMENT TYPE: Patent  
 LANGUAGE: English  
 FAMILY ACC. NUM. COUNT: 3  
 PATENT INFORMATION:

| PATENT NO.                                                                                                                                                                                                                                                                                                                                                                                                      | KIND                                                                                                                                                                                                     | DATE     | APPLICATION NO. | DATE       |
|-----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|----------|-----------------|------------|
| WO 2003002128                                                                                                                                                                                                                                                                                                                                                                                                   | A1                                                                                                                                                                                                       | 20030109 | WO 2002-FI575   | 20020628   |
| W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH,<br>CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES,<br>FI, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG,<br>KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW,<br>MX, MZ, NO                                                                                                                                           | RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, CH,<br>CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR,<br>BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG |          |                 |            |
| FI 2001001403                                                                                                                                                                                                                                                                                                                                                                                                   | A                                                                                                                                                                                                        | 20021230 | FI 2001-1403    | 20010629   |
| WO 2002056893                                                                                                                                                                                                                                                                                                                                                                                                   | A1                                                                                                                                                                                                       | 20020725 | WO 2002-FI43    | 20020118   |
| W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH,<br>CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES,<br>FI, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG,<br>KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW,<br>MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SK,<br>SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZM, ZW,<br>AM, AZ, BY, KG | RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, CH,<br>CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR,<br>BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG |          |                 |            |
| PRIORITY APPLN. INFO.:                                                                                                                                                                                                                                                                                                                                                                                          |                                                                                                                                                                                                          |          | FI 2001-1403    | A 20010629 |
|                                                                                                                                                                                                                                                                                                                                                                                                                 |                                                                                                                                                                                                          |          | WO 2002-FI43    | A 20020118 |
|                                                                                                                                                                                                                                                                                                                                                                                                                 |                                                                                                                                                                                                          |          | FI 2001-118     | A 20010119 |

AB The present invention relates to a compn. comprising a polysaccharide with Helicobacter pylori receptor activity and, optionally, an oligosaccharide receptor of Helicobacter pylori or an analog or a deriv. thereof and/or a gastric epithelium protecting compd. for use in the treatment or prophylaxis of any condition due to the presence of Helicobacter pylori. Binding assays revealed the isoreceptors and specificity of binding of glycolipids such as Neu5Gc.alpha.3Gal.beta.4GlcNAc.beta.3Gal.beta.4GlcNAc.beta.3Gal.beta.4Glc(beta).Cer.

REFERENCE COUNT: 17 THERE ARE 17 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L9 ANSWER 3 OF 6 CAPLUS COPYRIGHT 2003 ACS on STN DUPLICATE 1  
 ACCESSION NUMBER: 2002:764400 CAPLUS  
 DOCUMENT NUMBER: 138:233913  
 TITLE: Enzymatic Synthesis of Chondroitin with a Novel Chondroitin Sulfate N-Acetylgalactosaminyltransferase That Transfers N-Acetylgalactosamine to Glucuronic Acid in Initiation and Elongation of Chondroitin Sulfate Synthesis  
 AUTHOR(S): Gotoh, Masanori; Sato, Takashi; Akashima, Tomohiro;  
 Iwasaki, Hiroko; Kameyama, Akihiko; Mochizuki, Hideo;  
 Yada, Toshikazu; Inaba, Niro; Zhang, Yan; Kikuchi,  
 Norihiro; Kwon, Yeon-Dae; Togayachi, Akira; Kudo,  
 Takashi; Nishihara, Shoko; Watanabe, Hideto; Kimata,  
 Koji; Narimatsu, Hisashi  
 CORPORATE SOURCE: Glycogene Function Team, Res. Cent. Glycosci., Natl.  
 Inst. Adv. Ind. Sci. Technol., Ibaraki, 305-8586,  
 Japan  
 SOURCE: Journal of Biological Chemistry (2002), 277(41),  
 38189-38196  
 CODEN: JBCHA3; ISSN: 0021-9258

PUBLISHER: American Society for Biochemistry and Molecular Biology  
DOCUMENT TYPE: Journal  
LANGUAGE: English  
AB We found a novel glycosyltransferase gene having a hypothetical .beta.1,4-galactosyltransferase motif (GenBankTM accession no.) by a BLAST search and cloned its full-length open reading frame using the 5'-rapid amplification of cDNA ends method. The truncated form was expressed in insect cells as a sol. enzyme. It transferred N-acetylgalactosamine, not galactose, to para-nitrophenyl-.beta.-glucuronic acid. The N-acetylgalactosamine-glucuronic acid linkage has been identified only in chondroitin sulfate; therefore, we examd. its chondroitin elongation and initiation activities. N-Acetylgalactosaminyltransferase activity was obsd. toward chondroitin poly- and oligosaccharides, chondroitin sulfate oligosaccharides, and linkage tetrasaccharide (GlcA-Gal-Gal-Xyl-O-methoxyphenyl), and the chondroitin polysaccharide and linkage tetrasaccharide were better acceptor substrates than the others. Northern blot anal. and quant. real-time PCR anal. revealed that its 4-kb transcripts were highly expressed in thyroid and placenta, although they were ubiquitously expressed in various tissues and cells. These results suggest that this enzyme has N-acetylgalactosaminyltransferase activity in both the elongation and initiation of chondroitin sulfate synthesis. Furthermore, we performed enzymic synthesis of chondroitin pentasaccharide in vitro. In one tube reaction with four enzymes, .beta.1,4-galactosyltransferase-VII, .beta.1,3-galactosyltransferase-VI, glucuronyltransferase-I, and this enzyme, and a synthetic xylose-peptide acceptor, the structure GalNAc-GlcA-Gal-Gal-Xyl-peptide was constructed. This is the first report of a chondroitin pentasaccharide constructed with recombinant glycosyltransferases in vitro.

REFERENCE COUNT: 16 THERE ARE 16 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L9 ANSWER 4 OF 6 CAPLUS COPYRIGHT 2003 ACS on STN DUPLICATE 2  
ACCESSION NUMBER: 2001:480308 CAPLUS  
DOCUMENT NUMBER: 135:104525  
TITLE: Evidence of block and randomly sequenced chondroitin polysaccharides: Sequential enzymatic digestion and quantification using ion trap tandem mass spectrometry  
AUTHOR(S): Desaire, Heather; Sirich, Tammy L.; Leary, Julie A.  
CORPORATE SOURCE: College of Chemistry, University of California Berkeley, Berkeley, CA, 94720, USA  
SOURCE: Analytical Chemistry (2001), 73(15), 3513-3520  
CODEN: ANCHAM; ISSN: 0003-2700  
PUBLISHER: American Chemical Society  
DOCUMENT TYPE: Journal  
LANGUAGE: English  
AB A method for detg. the sequence type of the disaccharide repeat region of cartilage samples is introduced. The samples are sequentially subjected to selective and nonselective enzymic digestion, and the isomeric products from each step are quantified using tandem mass spectrometry. The two-step digestion/quantification protocol identifies whether the global makeup of the polymer is "alternating", "random", or "blocked" with respect to the two main components of the cartilage, 4- and 6-sulfated disaccharides. Using this procedure, the sequence type of two biol. isolated chondroitin polysaccharides was identified. The results for chondroitin sulfate A, isolated from bovine trachea, are consistent with the 4- and 6-sulfated disaccharides randomly distributed throughout the repeat region of the polysaccharide. For chondroitin sulfate C, shark cartilage, the 6-sulfated disaccharides are adjacent to each other to a larger extent than one would expect for a randomly distributed polymer, indicating that "blocks" of repeating disaccharides with the same sulfation site are present.

REFERENCE COUNT: 31 THERE ARE 31 CITED REFERENCES AVAILABLE FOR THIS

RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L9 ANSWER 5 OF 6 CAPLUS COPYRIGHT 2003 ACS on STN  
 ACCESSION NUMBER: 1995:277252 CAPLUS  
 DOCUMENT NUMBER: 122:38941  
 TITLE: Polysaccharide-immobilized substrates for removal of glycoproteins  
 INVENTOR(S): Shimizu, Yoshihiro  
 PATENT ASSIGNEE(S): Toray Industries, Japan  
 SOURCE: Jpn. Kokai Tokkyo Koho, 3 pp.  
 CODEN: JKXXAF  
 DOCUMENT TYPE: Patent  
 LANGUAGE: Japanese  
 FAMILY ACC. NUM. COUNT: 1  
 PATENT INFORMATION:

| PATENT NO.  | KIND | DATE     | APPLICATION NO. | DATE     |
|-------------|------|----------|-----------------|----------|
| JP 06261940 | A2   | 19940920 | JP 1993-53465   | 19930315 |
| JP 3271357  | B2   | 20020402 |                 |          |

PRIORITY APPLN. INFO.: JP 1993-53465 19930315  
 AB Carboxy-contg. polysaccharides are immobilized on substrates to be used for the removal of glycosylation products from the soln. For example, bovine serum albumin and glucose were dissolved in a phosphate buffer soln. and left at 37.degree. for 90 days. Addn. of a hyaluronate-immobilized EAH Sepharose 4B gel to the soln. resulted in a clear supernatant with a colored ptd. gel.

L9 ANSWER 6 OF 6 CAPLUS COPYRIGHT 2003 ACS on STN  
 ACCESSION NUMBER: 1969:26930 CAPLUS  
 DOCUMENT NUMBER: 70:26930  
 TITLE: Macromolecular polysaccharide proteins. III. Metabolic heterogeneity of chondroitin-4-sulfate-proteins and the metabolism of collagen in bovine cartilage  
 AUTHOR(S): Kresse, Hans; Buddecke, Eckhart  
 CORPORATE SOURCE: Univ. Muenster, Muenster, Fed. Rep. Ger.  
 SOURCE: Hoppe-Seyler's Zeitschrift fuer Physiologische Chemie (1968), 349(11), 1497-506  
 CODEN: HSZPAZ; ISSN: 0018-4888  
 DOCUMENT TYPE: Journal  
 LANGUAGE: German

AB Bovine nasal cartilage slices were incubated for 6 hrs. with labeled sulfate, glucose, glycine, or leucine. For at least 6 hrs. sulfate was incorporated into light protein polysaccharide, heavy protein polysaccharide, and protein polysaccharide residue, components of chondroitin sulfate protein, at different but const. rates. The specific radioactivities of the chondroitin sulfate from the 3 fractions were in the ratio of 1:1.8:0.7. After the incorporation of glucose, the specific radioactivities of these components were in the ratio of 1:4.9:1.3. The amino acids were incorporated into the protein components of light protein polysaccharide and urea-extd. heavy protein polysaccharide. The rate of incorporation of the amino acids into the protein components of the light protein polysaccharide and urea-treated heavy protein polysaccharide was 6-12 times higher than that into the collagen of bovine nasal cartilage. The specific radioactivity of the collagen fraction of heavy protein polysaccharide was 2-fold greater than that of the protein polysaccharide residue.

d his

(FILE 'HOME' ENTERED AT 15:11:28 ON 03 SEP 2003)

FILE 'CPLUS, MEDLINE' ENTERED AT 15:11:36 ON 03 SEP 2003

L1           0 S CHONDROITIN POLYMER  
L2           10 S CHONDROITIN POLYMER  
L3           0 S L2 AND COMPOSITION  
L4           7 S L2 AND CHONDROITIN SYNTHASE  
L5           3 S L2 AND SULFATED  
L6           2 DUP REM L5 (1 DUPLICATE REMOVED)  
L7           7 S L2 NOT L5  
L8           8 S CHONDROITIN POLYSACCHARIDE  
L9           6 DUP REM L8 (2 DUPLICATES REMOVED)

L12 ANSWER 11 OF 11 MEDLINE on STN  
ACCESSION NUMBER: 2000420900 MEDLINE  
DOCUMENT NUMBER: 20379058 PubMed ID: 10818104  
TITLE: Identification and molecular cloning of a  
chondroitin synthase from *Pasteurella*  
*multocida* type F.  
AUTHOR: DeAngelis P L; Padgett-McCue A J  
CORPORATE SOURCE: Department of Biochemistry and Molecular Biology,  
University of Oklahoma Health Sciences Center, Oklahoma  
City, Oklahoma 73104, USA.  
CONTRACT NUMBER: GM56497 (NIGMS)  
SOURCE: JOURNAL OF BIOLOGICAL CHEMISTRY, (2000 Aug 4) 275 (31)  
24124-9.  
Journal code: 2985121R.. ISSN: 0021-9258.  
PUB. COUNTRY: United States  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
OTHER SOURCE: GENBANK-AF195517  
ENTRY MONTH: 200009  
ENTRY DATE: Entered STN: 20000915  
Last Updated on STN: 20000915  
Entered Medline: 20000907  
AB *Pasteurella multocida* Type F, the minor fowl cholera pathogen, produces an extracellular polysaccharide capsule that is a putative virulence factor. It was reported that the capsule was removed by treating microbes with chondroitin AC lyase. We found by acid hydrolysis that the polysaccharide contained galactosamine and glucuronic acid. We molecularly cloned a Type F polysaccharide synthase and characterized its enzymatic activity. The 965-residue enzyme, called *P. multocida* chondroitin synthase (pmCS), is 87% identical at the nucleotide and the amino acid level to the hyaluronan synthase, pmHAS, from *P. multocida* Type A. A recombinant Escherichia coli-derived truncated, soluble version of pmCS (residues 1-704) was shown to catalyze the repetitive addition of sugars from UDP-GalNAc and UDP-GlcUA to chondroitin oligosaccharide acceptors in vitro. Other structurally related sugar nucleotide precursors did not substitute in the elongation reaction. Polymer molecules composed of approximately 10(3) sugar residues were produced, as measured by gel filtration chromatography. The polysaccharide synthesized in vitro was sensitive to the action of chondroitin AC lyase but resistant to the action of hyaluronan lyase. This is the first report identifying a glycosyltransferase that forms a polysaccharide composed of chondroitin disaccharide repeats, [ $\beta$ (1,4)GlcUA- $\beta$ (1,3)GalNAc]<sub>n</sub>. In analogy to known hyaluronan synthases, a single polypeptide species, pmCS, possesses both transferase activities.

L12 ANSWER 1 OF 11 CAPLUS COPYRIGHT 2003 ACS on STN  
ACCESSION NUMBER: 2003:478357 CAPLUS  
TITLE: Molecular Cloning of a Chondroitin  
Polymerizing Factor that Cooperates with  
Chondroitin Synthase for  
Chondroitin Polymerization  
AUTHOR(S): Kitagawa, Hiroshi; Izumikawa, Tomomi; Uyama, Toru;  
Sugahara, Kazuyuki  
CORPORATE SOURCE: Department of Biochemistry, Kobe Pharmaceutical  
University, Kobe, 658-8558, Japan  
SOURCE: Journal of Biological Chemistry (2003), 278(26),  
23666-23671  
CODEN: JBCHA3; ISSN: 0021-9258  
PUBLISHER: American Society for Biochemistry and Molecular  
Biology  
DOCUMENT TYPE: Journal  
LANGUAGE: English  
AB We recently cloned human chondroitin synthase (ChSy) exhibiting the glucuronyltransferase-II (GlcATII) and N-acetylgalactosaminyltransferase-II (GalNAcTII) activities responsible for the biosynthesis of repeating disaccharide units of chondroitin sulfate, but chondroitin polymn. was not demonstrated in vitro using the recombinant ChSy. We report here that the chondroitin polymg. activity requires concomitant expression of a novel protein designated chondroitin polymg. factor (ChPF) with ChSy. The human ChPF consists of 775 amino acids with a type II transmembrane protein topol. The amino acid sequence displayed 23% identity to that of human ChSy. The expression of a sol. recombinant form of the protein in COS-1 cells produced a protein with little GlcAT-II or GalNAcT-II activity. In contrast, coexpression of the ChPF and ChSy yielded markedly augmented glycosyltransferase activities, whereas simple mixing of the two sep. expressed proteins did not. Moreover, using both UDP-glucuronic acid (GlcUA) and UDP-N-acetylgalactosamine (GalNAc) as sugar donors, chondroitin polymn. was demonstrated on the so-called glycosaminoglycan-protein linkage region tetrasaccharide sequence of .alpha.-thrombomodulin. These results suggested that the ChPF acts as a specific activating factor for ChSy in chondroitin polymn. The coding region of the ChPF was divided into four discrete exons and localized to chromosome 2q35-q36. Northern blot anal. revealed that the ChPF gene exhibited a markedly different expression pattern among various human tissues, which was similar to that of ChSy. Thus, the ChPF is required for chondroitin polymg. activity of mammalian ChSy.  
REFERENCE COUNT: 30 THERE ARE 30 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L12 ANSWER 2 OF 11 CAPLUS COPYRIGHT 2003 ACS on STN  
ACCESSION NUMBER: 2003:443700 CAPLUS  
DOCUMENT NUMBER: 139:32505  
TITLE: Second human chondroitin  
N-acetylgalactosaminyltransferase chondroitin  
GalNAcT-2, encoding cDNA, and uses  
INVENTOR(S): Sugawara, Kazuyuki; Kitagawa, Hiroyuki  
PATENT ASSIGNEE(S): Shinsangyo Sozo Kenkyu Kiko, Japan  
SOURCE: Jpn. Kokai Tokkyo Koho, 35 pp.  
CODEN: JKXXAF  
DOCUMENT TYPE: Patent  
LANGUAGE: Japanese  
FAMILY ACC. NUM. COUNT: 1  
PATENT INFORMATION:

| PATENT NO. | KIND  | DATE  | APPLICATION NO. | DATE  |
|------------|-------|-------|-----------------|-------|
| -----      | ----- | ----- | -----           | ----- |

JP 2003164291 A2 20030610 JP 2001-367196 20011130  
 PRIORITY APPLN. INFO.: JP 2001-367196 20011130  
 AB A novel human chondroitin N-acetylgalactosaminyltransferase,  
 chondroitin N-acetylgalactosaminyltransferase 2 (chondroitin synthase 2), its encoding cDNA, recombinant sol. fusion protein, recombinant expression, and use as reagent for N-acetylgalactosamine-contg. sugar chain, are disclosed. Hybridization probes contg. sequence complementary to the gene sequence for chondroitin synthase 2, are claimed. The authors identified a novel human chondroitin N-acetylgalactosaminyltransferase, designated chondroitin GalNAcT-2 after a BLAST anal. of the GenBankTM data base using the sequence of a previously described human chondroitin N-acetylgalactosaminyltransferase (chondroitin GalNAcT-1) as a probe. The new cDNA sequence contained an open reading frame encoding a protein of 542 amino acids with a type II transmembrane protein topol. The amino acid sequence displayed 60% identity to that of human chondroitin GalNAcT-1. Like chondroitin GalNAcT-1, the expression of a sol. form of the protein in COS-1 cells produced an active enzyme, which not only transferred .beta.1,4-N-acetylgalactosamine (GalNAc) from UDP-[3H]GalNAc to a polymer chondroitin representing growing chondroitin chains (.beta.-GalNActransferase II activity) but also to GlcUA.beta.1-3Gal.beta.1-O-C2H4NHCbz, a synthetic substrate for .beta.-GalNActransferase I that transfers the first GalNAc to the core tetrasaccharide in the protein-linkage region of chondroitin sulfate. In contrast, the tetrasaccharide serine (GlcUA.beta.1-3Gal.beta.1-3Gal.beta.1-4Xyl.beta.1-O-Ser) derived from the linkage region, which is an inert acceptor substrate for chondroitin GalNAcT-1, served as an acceptor substrate. The coding region of this enzyme was divided into seven discrete exons, which is similar to the genomic organization of the chondroitin GalNAcT-1 gene, and was localized to chromosome 10q11.22. Northern blot anal. revealed that the chondroitin GalNAcT-2 gene exhibited a ubiquitous but differing expression in human tissues, and the expression pattern differed from that of chondroitin GalNAcT-1. Thus, the authors demonstrated redundancy in the chondroitin GalNActransferases involved in the biosynthetic initiation and elongation of chondroitin sulfate, which is important for understanding the biosynthetic mechanisms leading to the selective chain assembly of chondroitin/dermatan sulfate on the linkage region tetrasaccharide common to various proteoglycans contg. chondroitin/dermatan sulfate and heparin/heparan sulfate chains.

L12 ANSWER 3 OF 11 CAPLUS COPYRIGHT 2003 ACS on STN  
 ACCESSION NUMBER: 2001:798014 CAPLUS  
 DOCUMENT NUMBER: 135:354689  
 TITLE: Pasteurella multocida chondroitin synthase gene, recombinant expression, and use  
 INVENTOR(S): De Angelis, Paul L.  
 PATENT ASSIGNEE(S): USA  
 SOURCE: PCT Int. Appl., 125 pp.  
 CODEN: PIXXD2  
 DOCUMENT TYPE: Patent  
 LANGUAGE: English  
 FAMILY ACC. NUM. COUNT: 12  
 PATENT INFORMATION:

| PATENT NO.                                                                                                                         | KIND | DATE     | APPLICATION NO. | DATE     |
|------------------------------------------------------------------------------------------------------------------------------------|------|----------|-----------------|----------|
| WO 2001080810                                                                                                                      | A2   | 20011101 | WO 2001-US13395 | 20010425 |
| WO 2001080810                                                                                                                      | A3   | 20020516 |                 |          |
| W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, |      |          |                 |          |

HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS,  
LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO,  
RU, SD, SE

RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY,  
DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF,  
BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG

US 6444447 B1 20020903 US 1999-437277 19991110

AU 2001053805 A5 20011107 AU 2001-53805 20010425

EP 1282684 A2 20030212 EP 2001-927344 20010425

R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT,  
IE, SI, LT, LV, FI, RO, MK, CY, AL, TR

PRIORITY APPLN. INFO.: US 1999-437277 A2 19991110

US 1998-107929P P 19981111

US 1999-283402 A2 19990401

WO 2001-US13395 W 20010425

AB The present invention relates to a **chondroitin synthase** gene from *Pasteurella multocida* and methods of isolating, recombinant expression, and use in unsulfated **chondroitin** prepns. Addnl., the present invention relates to the use of unsulfated **chondroitin** and its prepns., as well as conversion into modified versions such as dermatan sulfate and **chondroitin sulfate** polymers. *Pasteurella multocida* Type F, the minor fowl cholera pathogen, produces an extracellular polysaccharide capsule that is a putative virulence factor. It was reported that the capsule was removed by treating microbes with **chondroitin AC lyase**. We found by acid hydrolysis that the polysaccharide contained galactosamine and glucuronic acid. We molecularly cloned a Type F polysaccharide synthase and characterized its enzymic activity. The 965-residue enzyme, called *P. multocida chondroitin synthase* (pmCS), is 87% identical at the nucleotide and the amino acid level to the hyaluronan synthase, pmHAS, from *P. multocida* Type A. A recombinant *Escherichia coli*-derived truncated, sol. version of pmCS (residues 1-704) was shown to catalyze the repetitive addn. of sugars from UDP-GalNAc and UDP-GlcUA to **chondroitin oligosaccharide acceptors** in vitro. Other structurally related sugar nucleotide precursors did not substitute in the elongation reaction. Polymer mols. composed of .apprx.103 sugar residues were produced, as measured by gel filtration chromatog. The polysaccharide synthesized in vitro was sensitive to the action of **chondroitin AC lyase** but resistant to the action of hyaluronan lyase. This is the first report identifying a glycosyltransferase that forms a polysaccharide composed of **chondroitin disaccharide repeats**, [.beta.(1,4)GlcUA-.beta.(1,3)GalNAc]n. In analogy to known hyaluronan synthases, a single polypeptide species, pmCS, possesses both transferase activities. Directly downstream of the pmCS gene a putative UDP-glucose dehydrogenase gene was identified.

L12 ANSWER 4 OF 11 CAPLUS COPYRIGHT 2003 ACS on STN

ACCESSION NUMBER: 2001:783462 CAPLUS

DOCUMENT NUMBER: 136:289757

TITLE: Molecular cloning and expression of a human  
**chondroitin synthase**

AUTHOR(S): Kitagawa, Hiroshi; Uyama, Toru; Sugahara, Kazuyuki

CORPORATE SOURCE: Department of Biochemistry, Kobe Pharmaceutical  
University, Kobe, 658-8558, Japan

SOURCE: Journal of Biological Chemistry (2001), 276(42),  
38721-38726

CODEN: JBCHA3; ISSN: 0021-9258

PUBLISHER: American Society for Biochemistry and Molecular  
Biology

DOCUMENT TYPE: Journal

LANGUAGE: English

AB We have identified a human **chondroitin synthase** from the HUGE (human unidentified gene-encoded large proteins) protein data base by screening with two keywords: "one transmembrane domain" and

"galactosyltransferase family.". The identified protein consists of 802 amino acids with a type II transmembrane protein topol. The protein showed weak homol. to the .beta.1,3-galactosyltransferase family on the amino-terminal side and to the .beta.1,4-galactosyltransferase family on the carboxyl-terminal side. The expression of a sol. recombinant form of the protein in COS-1 cells produced an active enzyme, which transferred not only the glucuronic acid (GlcUA) from UDP-[14C]GlcUA but also N-acetylgalactosamine (GalNAc) from UDP-[3H]GalNAc to the polymer chondroitin. Identification of the reaction products demonstrated that the enzyme was chondroitin synthase, with both .beta.1,3-GlcUA transferase and .beta.1,4-GalNAc transferase activities. The coding region of the chondroitin synthase was divided into three discrete exons and localized to chromosome 15. Northern blot anal. revealed that the chondroitin synthase gene exhibited ubiquitous but markedly differential expression in the human tissues examd. Thus, we demonstrated that analogous to human heparan sulfate polymerases, the single polypeptide chondroitin synthase possesses two glycosyltransferase activities required for chain polymn.

REFERENCE COUNT: 41 THERE ARE 41 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L12 ANSWER 5 OF 11 CAPLUS COPYRIGHT 2003 ACS on STN

ACCESSION NUMBER: 2000:569635 CAPLUS

DOCUMENT NUMBER: 133:292768

TITLE: Identification and molecular cloning of a chondroitin synthase from Pasteurella multocida type F

AUTHOR(S): DeAngelis, Paul L.; Padgett-McCue, Amy J.

CORPORATE SOURCE: Department of Biochemistry and Molecular Biology, University of Oklahoma Health Sciences Center, Oklahoma City, OK, 73104, USA

SOURCE: Journal of Biological Chemistry (2000), 275(31), 24124-24129

CODEN: JBCHA3; ISSN: 0021-9258

PUBLISHER: American Society for Biochemistry and Molecular Biology

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Pasteurella multocida Type F, the minor fowl cholera pathogen, produces an extracellular polysaccharide capsule that is a putative virulence factor. It was reported that the capsule was removed by treating microbes with chondroitin AC lyase. We found by acid hydrolysis that the polysaccharide contained galactosamine and glucuronic acid. We molecularly cloned a Type F polysaccharide synthase and characterized its enzymic activity. The 965-residue enzyme, called P. multocida chondroitin synthase (pmCS), is 87% identical at the nucleotide and the amino acid level to the hyaluronan synthase, pmHAS, from P. multocida Type A. A recombinant Escherichia coli-derived truncated, sol. version of pmCS (residues 1-704) was shown to catalyze the repetitive addn. of sugars from UDP-GalNAc and UDP-GlcUA to chondroitin oligosaccharide acceptors in vitro. Other structurally related sugar nucleotide precursors did not substitute in the elongation reaction. Polymer mols. composed of .apprx.10<sup>3</sup> sugar residues were produced, as measured by gel filtration chromatog. The polysaccharide synthesized in vitro was sensitive to the action of chondroitin AC lyase but resistant to the action of hyaluronan lyase. This is the first report identifying a glycosyltransferase that forms a polysaccharide composed of chondroitin disaccharide repeats, [.beta.(1,4)GlcUA-.beta.(1,3)GalNAc]<sub>n</sub>. In analogy to known hyaluronan synthases, a single polypeptide species, pmCS, possesses both transferase activities.

REFERENCE COUNT: 33 THERE ARE 33 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L12 ANSWER 6 OF 11 CAPLUS COPYRIGHT 2003 ACS on STN  
 ACCESSION NUMBER: 1999:659266 CAPLUS  
 DOCUMENT NUMBER: 131:296218  
 TITLE: Nucleic acid encoding hyaluronan synthase from  
       *Pasteurella multocida*  
 INVENTOR(S): Deangelis, Paul  
 PATENT ASSIGNEE(S): Board of Regents of the University of Oklahoma, USA  
 SOURCE: PCT Int. Appl., 121 pp.  
 CODEN: PIXXD2  
 DOCUMENT TYPE: Patent  
 LANGUAGE: English  
 FAMILY ACC. NUM. COUNT: 12  
 PATENT INFORMATION:

| PATENT NO.                                                                                                                                                                                                                                                                                                                                                   | KIND | DATE     | APPLICATION NO. | DATE        |
|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|------|----------|-----------------|-------------|
| WO 9951265                                                                                                                                                                                                                                                                                                                                                   | A1   | 19991014 | WO 1999-US7289  | 19990401    |
| W: AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ,<br>DE, DK, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS,<br>JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK,<br>MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ,<br>TM, TR, TT, UA, UG, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD,<br>RU, TJ, TM |      |          |                 |             |
| RW: GH, GM, KE, LS, MW, SD, SL, SZ, UG, ZW, AT, BE, CH, CY, DE, DK,<br>ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG,<br>CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG                                                                                                                                                                         |      |          |                 |             |
| CA 2326821                                                                                                                                                                                                                                                                                                                                                   | AA   | 19991014 | CA 1999-2326821 | 19990401    |
| AU 9935485                                                                                                                                                                                                                                                                                                                                                   | A1   | 19991025 | AU 1999-35485   | 19990401    |
| EP 1073460                                                                                                                                                                                                                                                                                                                                                   | A1   | 20010207 | EP 1999-917339  | 19990401    |
| R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT,<br>IE, SI, LT, LV, FI, RO                                                                                                                                                                                                                                                                 |      |          |                 |             |
| BR 9909346                                                                                                                                                                                                                                                                                                                                                   | A    | 20010911 | BR 1999-9346    | 19990401    |
| JP 2002510648                                                                                                                                                                                                                                                                                                                                                | T2   | 20020409 | JP 2000-542035  | 19990401    |
| US 2003113845                                                                                                                                                                                                                                                                                                                                                | A1   | 20030619 | US 2002-217613  | 20020812    |
| PRIORITY APPLN. INFO.:                                                                                                                                                                                                                                                                                                                                       |      |          | US 1998-80414P  | P 19980402  |
|                                                                                                                                                                                                                                                                                                                                                              |      |          | US 1998-178851  | A 19981026  |
|                                                                                                                                                                                                                                                                                                                                                              |      |          | US 1999-283402  | B1 19990401 |
|                                                                                                                                                                                                                                                                                                                                                              |      |          | WO 1999-US7289  | W 19990401  |

AB The present invention relates to a nucleic acid segment having a coding region segment encoding enzymically active hyaluronate synthase (PmHAS) from fowl cholera pathogen Type A *Pasteurella multocida*, and to the use of this nucleic acid segment in the prepn. of recombinant cells which produce hyaluronate synthase and its hyaluronic acid product. Hyaluronate is also known as hyaluronic acid or hyaluronan. Sections of the first 420 residues of PmHAS show some similarity to portions of mammalian UDP-GalNAc:polypeptide N-acetylgalactosaminyltransferase, but the last apprx. 340 residues of PmHAS are not significantly similar to other entries in the sequence databases. Thus, *P. multocida* HAS is unique and most likely the prototype of an entire new class of HAS. The measured Km values for PmHAS in crude membranes are about 2-3-fold lower for UDP-GlcNAc and UDP-GlcA than those obtained from HAS found in streptococcal membranes. A homologous gene for Type F *Pasteurella multocida chondroitin synthase* is also provided. The present invention also relates to the use of the PmHAS in constructing "knock-out" mutant strains of *P. multocida* for use in vaccinations. The present invention further relates to the use of the PmHAS in diagnostic tests in the field detns. of livestock *P. multocida* infection.

REFERENCE COUNT: 8 THERE ARE 8 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L12 ANSWER 7 OF 11 MEDLINE on STN  
 ACCESSION NUMBER: 2003292145 MEDLINE  
 DOCUMENT NUMBER: 22703888 PubMed ID: 12716890

TITLE: Molecular cloning of a **chondroitin** polymerizing factor that cooperates with **chondroitin synthase** for **chondroitin** polymerization.

AUTHOR: Kitagawa Hiroshi; Izumikawa Tomomi; Uyama Toru; Sugahara Kazuyuki

CORPORATE SOURCE: Department of Biochemistry, Kobe Pharmaceutical University, Higashinada-ku, Kobe 658-8558, Japan.

SOURCE: JOURNAL OF BIOLOGICAL CHEMISTRY, (2003 Jun 27) 278 (26) 23666-71.  
Journal code: 2985121R. ISSN: 0021-9258.

PUB. COUNTRY: United States

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

OTHER SOURCE: GENBANK-AB095813

ENTRY MONTH: 200308

ENTRY DATE: Entered STN: 20030624  
Last Updated on STN: 20030821  
Entered Medline: 20030820

AB We recently cloned human **chondroitin synthase** (ChSy) exhibiting the glucuronyltransferase-II (GlcATII) and N-acetylgalactosaminyltransferase-II (GalNAcTII) activities responsible for the biosynthesis of repeating disaccharide units of **chondroitin** sulfate, but **chondroitin** polymerization was not demonstrated in vitro using the **recombinant** ChSy. We report here that the **chondroitin** polymerizing activity requires concomitant expression of a novel protein designated **chondroitin** polymerizing factor (ChPF) with ChSy. The human ChPF consists of 775 amino acids with a type II transmembrane protein topology. The amino acid sequence displayed 23% identity to that of human ChSy. The expression of a soluble **recombinant** form of the protein in COS-1 cells produced a protein with little GlcAT-II or GalNAcT-II activity. In contrast, coexpression of the ChPF and ChSy yielded markedly augmented glycosyltransferase activities, whereas simple mixing of the two separately expressed proteins did not. Moreover, using both UDP-glucuronic acid (GlcUA) and UDP-N-acetylgalactosamine (GalNAc) as sugar donors, **chondroitin** polymerization was demonstrated on the so-called glycosaminoglycan-protein linkage region tetrasaccharide sequence of alpha-thrombomodulin. These results suggested that the ChPF acts as a specific activating factor for ChSy in **chondroitin** polymerization. The coding region of the ChPF was divided into four discrete exons and localized to chromosome 2q35-q36. Northern blot analysis revealed that the ChPF gene exhibited a markedly different expression pattern among various human tissues, which was similar to that of ChSy. Thus, the ChPF is required for **chondroitin** polymerizing activity of mammalian ChSy.

L12 ANSWER 8 OF 11 MEDLINE on STN

ACCESSION NUMBER: 2002620027 MEDLINE

DOCUMENT NUMBER: 22254828 PubMed ID: 12145278

TITLE: Molecular cloning and characterization of a novel **chondroitin** sulfate glucuronyltransferase that transfers glucuronic acid to N-acetylgalactosamine.

AUTHOR: Gotoh Masanori; Yada Toshikazu; Sato Takashi; Akashima Tomohiro; Iwasaki Hiroko; Mochizuki Hideo; Inaba Niro; Togayachi Akira; Kudo Takashi; Watanabe Hideto; Kimata Koji; Narimatsu Hisashi

CORPORATE SOURCE: Glycogene Function Team, Research Center for Glycoscience, National Institute of Advanced Industrial Science and Technology, Open Space Laboratory, C-2, 1-1-1 Umezono, Tsukuba, Ibaraki 305-8568, Japan.

SOURCE: JOURNAL OF BIOLOGICAL CHEMISTRY, (2002 Oct 11) 277 (41) 38179-88.  
Journal code: 2985121R. ISSN: 0021-9258.

PUB. COUNTRY: United States

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
OTHER SOURCE: GENBANK-AB037823  
ENTRY MONTH: 200211  
ENTRY DATE: Entered STN: 20021017  
Last Updated on STN: 20030105  
Entered Medline: 20021125

AB We found a novel human gene (GenBank accession number, Kazusa DNA Research Institute KIAA1402) that possesses homology with **chondroitin synthase**. The full-length open reading frame consists of 772 amino acids and encodes a typical type II membrane protein. This enzyme had a domain containing beta 3-glycosyltransferase motifs, which might be a beta3-glucuronyltransferase domain, but no domain with beta 4-glycosyltransferase motifs, although both are found in **chondroitin synthase**. The putative catalytic domain was expressed in COS-7 cells as a soluble enzyme. Its glucuronyltransferase activity was observed when **chondroitin** and **chondroitin sulfate** polysaccharides and oligosaccharides were used as acceptor substrates. However, it was not detected when dermatan sulfate, hyaluronan, heparan sulfate, heparin, N-acetylheparosan, lactosamine tetrasaccharide, and linkage tri- and tetrasaccharide acceptors were employed. The reaction product, which was speculated to exhibit a GlcA beta 1-3GalNAc linkage structure at its non-reducing terminus, showed the following characteristics. 1) It was catabolized by beta-glucuronidase. 2) It was an acceptor for Escherichia coli K4 **chondroitin polymerase** (K4 **chondroitin polymerase**). 3) The product of K4 **chondroitin polymerase** was cleaved by chondroitinase ACII. On the other hand, no N-acetylgalactosaminyltransferase activity was detected toward any acceptors. Quantitative real time PCR analysis revealed that its transcripts were highly expressed in the placenta, small intestine, and pancreas, although they were ubiquitously expressed in various tissues and cell lines. This enzyme could play a role in the synthesis of **chondroitin sulfate** as a glucuronyltransferase.

L12 ANSWER 9 OF 11 MEDLINE on STN  
ACCESSION NUMBER: 2002330828 MEDLINE  
DOCUMENT NUMBER: 22050655 PubMed ID: 11943778  
TITLE: Molecular cloning and characterization of **chondroitin polymerase** from Escherichia coli strain K4.  
AUTHOR: Ninomiya Toshio; Sugiura Nobuo; Tawada Akira; Sugimoto Kazunori; Watanabe Hideto; Kimata Koji  
CORPORATE SOURCE: Institute for Molecular Science of Medicine, Aichi Medical University, Yazako, Nagakute, Aichi 480-1195, Japan.  
SOURCE: JOURNAL OF BIOLOGICAL CHEMISTRY, (2002 Jun 14) 277 (24) 21567-75.  
Journal code: 2985121R. ISSN: 0021-9258.  
PUB. COUNTRY: United States  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
OTHER SOURCE: GENBANK-AB079602; GENBANK-X77617  
ENTRY MONTH: 200207  
ENTRY DATE: Entered STN: 20020621  
Last Updated on STN: 20030105  
Entered Medline: 20020725

AB Escherichia coli strain K4 produces the K4 antigen, a capsule polysaccharide consisting of a **chondroitin** backbone (GlcUA beta(1-3)-GalNAc beta(1-4))(n) to which beta-fructose is linked at position C-3 of the GlcUA residue. We molecularly cloned region 2 of the K4 capsular gene cluster essential for biosynthesis of the polysaccharide, and we further identified a gene encoding a bifunctional glycosyltransferase that polymerizes the **chondroitin** backbone.

The enzyme, containing two conserved glycosyltransferase sites, showed 59 and 61% identity at the amino acid level to class 2 hyaluronan synthase and **chondroitin synthase** from *Pasteurella multocida*, respectively. The soluble enzyme expressed in a bacterial expression system transferred GalNAc and GlcUA residues alternately, and polymerized the **chondroitin** chain up to a molecular mass of 20 kDa when **chondroitin sulfate hexasaccharide** was used as an acceptor. The enzyme exhibited apparent K(m) values for UDP-GlcUA and UDP-GalNAc of 3.44 and 31.6 microm, respectively, and absolutely required acceptors of **chondroitin sulfate polymers** and oligosaccharides at least longer than a tetrasaccharide. In addition, **chondroitin polymers** and oligosaccharides and hyaluronan polymers and oligosaccharides served as acceptors for **chondroitin** polymerization, but dermatan sulfate and heparin did not. These results may lead to elucidation of the mechanism for **chondroitin** chain synthesis in both microorganisms and mammals.

L12 ANSWER 10 OF 11 MEDLINE on STN  
ACCESSION NUMBER: 2001553876 MEDLINE  
DOCUMENT NUMBER: 21486534 PubMed ID: 11514575  
TITLE: Molecular cloning and expression of a human  
**chondroitin synthase**.  
AUTHOR: Kitagawa H; Uyama T; Sugahara K  
CORPORATE SOURCE: Department of Biochemistry, Kobe Pharmaceutical University,  
Higashinada-ku, Kobe 658-8558, Japan.  
SOURCE: JOURNAL OF BIOLOGICAL CHEMISTRY, (2001 Oct 19) 276 (42)  
38721-6.  
Journal code: 2985121R. ISSN: 0021-9258.  
PUB. COUNTRY: United States  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 200112  
ENTRY DATE: Entered STN: 20011016  
Last Updated on STN: 20030105  
Entered Medline: 20011204

AB We have identified a human **chondroitin synthase** from the HUGE (human unidentified gene-encoded large proteins) protein data base by screening with two keywords: "one transmembrane domain" and "galactosyltransferase family." The identified protein consists of 802 amino acids with a type II transmembrane protein topology. The protein showed weak homology to the beta1,3-galactosyltransferase family on the amino-terminal side and to the beta1,4-galactosyltransferase family on the carboxyl-terminal side. The expression of a soluble recombinant form of the protein in COS-1 cells produced an active enzyme, which transferred not only the glucuronic acid (GlcUA) from UDP-[<sup>14</sup>C]GlcUA but also N-acetylgalactosamine (GalNAc) from UDP-[<sup>3</sup>H]GalNAc to the polymer **chondroitin**. Identification of the reaction products demonstrated that the enzyme was **chondroitin synthase**, with both beta1,3-GlcUA transferase and beta1,4-GalNAc transferase activities. The coding region of the **chondroitin synthase** was divided into three discrete exons and localized to chromosome 15. Northern blot analysis revealed that the **chondroitin synthase** gene exhibited ubiquitous but markedly differential expression in the human tissues examined. Thus, we demonstrated that analogous to human heparan sulfate polymerases, the single polypeptide **chondroitin synthase** possesses two glycosyltransferase activities required for chain polymerization.

L12 ANSWER 11 OF 11 MEDLINE on STN  
ACCESSION NUMBER: 2000420900 MEDLINE  
DOCUMENT NUMBER: 20379058 PubMed ID: 10818104  
TITLE: Identification and molecular cloning of a  
**chondroitin synthase** from *Pasteurella*

AUTHOR: multocida type F.  
DeAngelis P L; Padgett-McCue A J  
CORPORATE SOURCE: Department of Biochemistry and Molecular Biology,  
University of Oklahoma Health Sciences Center, Oklahoma  
City, Oklahoma 73104, USA.  
CONTRACT NUMBER: GM56497 (NIGMS)  
SOURCE: JOURNAL OF BIOLOGICAL CHEMISTRY, (2000 Aug 4) 275 (31)  
24124-9.  
Journal code: 2985121R. ISSN: 0021-9258.  
PUB. COUNTRY: United States  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
OTHER SOURCE: GENBANK-AF195517  
ENTRY MONTH: 200009  
ENTRY DATE: Entered STN: 20000915  
Last Updated on STN: 20000915  
Entered Medline: 20000907

AB Pasteurella multocida Type F, the minor fowl cholera pathogen, produces an extracellular polysaccharide capsule that is a putative virulence factor. It was reported that the capsule was removed by treating microbes with chondroitin AC lyase. We found by acid hydrolysis that the polysaccharide contained galactosamine and glucuronic acid. We molecularly cloned a Type F polysaccharide synthase and characterized its enzymatic activity. The 965-residue enzyme, called *P. multocida chondroitin synthase* (pmCS), is 87% identical at the nucleotide and the amino acid level to the hyaluronan synthase, pmHAS, from *P. multocida* Type A. A recombinant Escherichia coli-derived truncated, soluble version of pmCS (residues 1-704) was shown to catalyze the repetitive addition of sugars from UDP-GalNAc and UDP-GlcUA to chondroitin oligosaccharide acceptors in vitro. Other structurally related sugar nucleotide precursors did not substitute in the elongation reaction. Polymer molecules composed of approximately 10(3) sugar residues were produced, as measured by gel filtration chromatography. The polysaccharide synthesized in vitro was sensitive to the action of chondroitin AC lyase but resistant to the action of hyaluronan lyase. This is the first report identifying a glycosyltransferase that forms a polysaccharide composed of chondroitin disaccharide repeats, [ $\beta$ (1,4)GlcUA- $\beta$ (1,3)GalNAc]<sub>n</sub>. In analogy to known hyaluronan synthases, a single polypeptide species, pmCS, possesses both transferase activities.

d his

(FILE 'HOME' ENTERED AT 15:11:28 ON 03 SEP 2003)

FILE 'CAPLUS, MEDLINE' ENTERED AT 15:11:36 ON 03 SEP 2003

L1           0 S CHRONDROITIN POLYMER  
L2           10 S CHONDROITIN POLYMER  
L3           0 S L2 AND COMPOSITION  
L4           7 S L2 AND CHONDROITIN SYNTHASE  
L5           3 S L2 AND SULFATED  
L6           2 DUP REM L5 (1 DUPLICATE REMOVED)  
L7           7 S L2 NOT L5  
L8           8 S CHONDROITIN POLYSACCHARIDE  
L9           6 DUP REM L8 (2 DUPLICATES REMOVED)  
L10          23094 S CHONDROITIN  
L11          35 S L10 AND CHONDROITIN SYNTHASE  
L12          11 S L11 AND RECOMBINANT  
L13          24 S L11 NOT L12  
L14          10 S L13 AND PASTEURELLA

L17 ANSWER 5 OF 5 MEDLINE on STN  
ACCESSION NUMBER: 85203849 MEDLINE  
DOCUMENT NUMBER: 85203849 PubMed ID: 3922754  
TITLE: Two N-acetylgalactosaminyltransferase are involved in the biosynthesis of chondroitin sulfate.  
AUTHOR: Rohrmann K; Niemann R; Buddecke E  
SOURCE: EUROPEAN JOURNAL OF BIOCHEMISTRY, (1985 May 2) 148 (3) 463-9.  
PUB. COUNTRY: Journal code: 0107600. ISSN: 0014-2956.  
DOCUMENT TYPE: GERMANY, WEST: Germany, Federal Republic of  
Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 198507  
ENTRY DATE: Entered STN: 19900320  
Last Updated on STN: 19900320  
Entered Medline: 19850705

AB Two N-acetylgalactosaminyltransferases, designated I and II, have been purified from the microsomal fraction of calf arterial tissue and separated on Bio-Gel A. N-Acetylgalactosaminyltransferase I was purified 450-fold. It requires Mn<sup>2+</sup> for maximal activity and transfers N-acetylgalactosamine residues from UDP-[1-3H]GalNAc in beta-glycosidic configuration to the non-reducing terminus of the acceptor substrates GlcA(beta 1-3)Gal(beta 1-3)Gal, GlcA(beta 1-3)Gal(beta 1-4)Glc and GlcA(beta 1-3)Gal. Even-numbered chondroitin oligosaccharides serve as acceptors for N-acetylgalactosaminyltransferase II, which transfers N-acetylgalactosamine from UDP-[1-3H]GalNAc to the non-reducing glucuronic acid residues of oligosaccharide acceptor substrates. Maximum transfer rates were obtained with a decasaccharide derived from chondroitin. Longer or shorter-chain chondroitin oligosaccharides are less effective acceptor substrates. All reaction products formed by N-acetylgalactosaminyltransferases I and II are substrates of beta-N-acetylhexosaminidase, which splits off the transferred [1-3H]GalNAc completely. In the microsomal fraction N-acetylgalactosaminyltransferase II had a 300-fold higher specific activity than N-acetylgalactosaminyltransferase I. In contrast to enzyme I, enzyme II loses much of its activity during the purification procedure and undergoes rapid thermodenaturation. GlcA-Gal-Gal is a characteristic sequence of the carbohydrate-protein linkage region of proteochondroitin sulfate. The acceptor capacity of this trisaccharide suggests that N-acetylgalactosaminyltransferase I is involved in the synthesis of the carbohydrate-protein linkage region. Since N-acetylgalactosaminyltransferase II is highly specific for chondroitin oligosaccharides, we conclude that it participates in chain elongation during chondroitin sulfate synthesis.

L17 ANSWER 4 OF 5 MEDLINE on STN  
ACCESSION NUMBER: 1999421651 MEDLINE  
DOCUMENT NUMBER: 99421651 PubMed ID: 10491092  
TITLE: Purification and characterization of fetal bovine serum  
beta-N-acetyl-D-galactosaminyltransferase and  
beta-D-glucuronyltransferase involved in chondroitin  
sulfate biosynthesis.  
AUTHOR: Tsuchida K; Lind T; Kitagawa H; Lindahl U; Sugahara K;  
Lidholt K  
CORPORATE SOURCE: Department of Biochemistry, Kobe Pharmaceutical University,  
Japan.  
SOURCE: EUROPEAN JOURNAL OF BIOCHEMISTRY, (1999 Sep) 264 (2) 461-7.  
Journal code: 0107600. ISSN: 0014-2956.  
PUB. COUNTRY: GERMANY: Germany, Federal Republic of  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 199910  
ENTRY DATE: Entered STN: 19991026  
Last Updated on STN: 19991026  
Entered Medline: 19991014

AB beta-N-Acetylgalactosaminyltransferase II and beta-glucuronyltransferase  
II, involved in chondroitin sulfate biosynthesis, transfer an  
N-acetylgalactosamine (GalNAc) and glucuronic acid (GlcA) residue,  
respectively, through beta-linkages to an acceptor **chondroitin**  
**oligosaccharide** derived from the repeating disaccharide region of  
chondroitin sulfate. They were copurified from fetal bovine serum  
approximately 2500-fold and 850-fold, respectively, by sequential  
chromatographies on Red A-agarose, phenyl-Sepharose, S-Sepharose and wheat  
germ agglutinin-agarose. Identical and inseparable chromatographic  
profiles of both glycosyltransferase activities obtained through the above  
chromatographic steps and gel filtration suggest that the **purified**  
enzyme activities are tightly coupled, which could imply a single enzyme  
with dual transferase activities; beta-N-acetylgalactosaminyltransferase  
and beta-glucuronyltransferase, reminiscent of the heparan sulfate  
polymerase reaction. However, when a polymerization reaction was  
performed in vitro with the **purified** serum enzyme preparation  
under the polymerization conditions recently developed for the  
chondroitin-synthesizing system, derived from human melanoma cells, each  
monosaccharide transfer took place, but no polymerization occurred. These  
results may suggest that the **purified** serum enzyme preparation  
contains both beta-N-acetylgalactosaminyltransferase II and  
beta-glucuronyltransferase II activities on a single polypeptide or on the  
respective polypeptides forming an enzyme complex, but is different from  
that obtained from melanoma cells in that it transfers a single GalNAc or  
GlcA residue but does not polymerize chondroitin.

L17 ANSWER 3 OF 5 CAPLUS COPYRIGHT 2003 ACS on STN  
ACCESSION NUMBER: 1981:116374 CAPLUS  
DOCUMENT NUMBER: 94:116374  
TITLE: Preparation and circular dichroism analysis of sodium hyaluronate oligosaccharides and chondroitin  
AUTHOR(S): Cowman, Mary K.; Balazs, Endre A.; Bergmann, Carl W.; Meyer, Karl  
CORPORATE SOURCE: Coll. Physicians Surg., Columbia Univ., New York, NY, 10032, USA  
SOURCE: Biochemistry (1981), 20(5), 1379-85  
CODEN: BICHAW; ISSN: 0006-2960  
DOCUMENT TYPE: Journal  
LANGUAGE: English

AB Na hyaluronate was cleaved into a homologous series of oligosaccharides by the action of bovine testicular hyaluronidase (EC 3.2.1.35), an endo-.beta.-hexosaminidase. Digestion conditions and gel-filtration chromatog. fractionation were optimized to produce oligosaccharides, clearly separable into peaks corresponding to 1-23 disaccharide units of the type D-glucuronosyl-N-acetyl-D-glucosamine. The chromatog. method was also employed for the purifn. of a 2nd homologous group of oligosaccharides, with the reversed sequence of monosaccharide units, produced by the action of leech hyaluronidase (EC 3.2.1.36), an endo-.beta.-glucuronidase. CD anal. (in the 200-250-nm range) of the oligosaccharides showed that the CD spectrum of hyaluronate in aq. soln. at neutral pH does not reflect to any substantial degree a polymer conformation which requires cooperative interaction between several repeating residues for stabilization. The enhanced CD properties of hyaluronate relative to monosaccharides are primarily related to the existence of the .beta.-1,4 linkage from N-acetyl-D-glucosamine to D-glucuronate. Chondroitin, the N-acetyl-D-galactosamine analog of hyaluronate, was prep'd. by chem. desulfation of chondroitin 4- and 6-sulfates. The **purified** product had a mol. wt. range of 4000-8000 (10-20 disaccharide units). It was digested with testicular hyaluronidase, and the split products were isolated by gel filtration. In contrast to hyaluronate, the cleavage products included both the preponderant analogous repeating disaccharide multiples with N-acetyl-D-galactosamine at the reducing end and smaller quantities of oligosaccharides composed of an odd no. of monosaccharides. These latter products were presumably derived from the ends of the shortened chondroitin chains, resulting from methanolysis during the desulfation and alk. elimination of reducing hexosamines. CD spectroscopic anal. of chondroitin relative to its constituent monosaccharides showed that this glycosaminoglycan does not share the structural feature which results in substantially enhanced CD properties for hyaluronate. A hypothesis relating the CD properties of N-acetylated hexosamines in oligo- and polysaccharides to the dihedral angle about the C-O bond at C3 may explain the CD dependence on both the hexosaminidic linkage and hexosamine configuration at C4.

L17 ANSWER 3 OF 5 CAPLUS COPYRIGHT 2003 ACS on STN

ACCESSION NUMBER: 1981:116374 CAPLUS

DOCUMENT NUMBER: 94:116374

TITLE: Preparation and circular dichroism analysis of sodium hyaluronate oligosaccharides and chondroitin

AUTHOR(S): Cowman, Mary K.; Balazs, Endre A.; Bergmann, Carl W.; Meyer, Karl

CORPORATE SOURCE: Coll. Physicians Surg., Columbia Univ., New York, NY, 10032, USA

SOURCE: Biochemistry (1981), 20(5), 1379-85  
CODEN: BICHAW; ISSN: 0006-2960

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Na hyaluronate was cleaved into a homologous series of oligosaccharides by the action of bovine testicular hyaluronidase (EC 3.2.1.35), an endo-.beta.-hexosaminidase. Digestion conditions and gel-filtration chromatog. fractionation were optimized to produce oligosaccharides, clearly separable into peaks corresponding to 1-23 disaccharide units of the type D-glucuronosyl-N-acetyl-D-glucosamine. The chromatog. method was also employed for the purifn. of a 2nd homologous group of oligosaccharides, with the reversed sequence of monosaccharide units, produced by the action of leech hyaluronidase (EC 3.2.1.36), an endo-.beta.-glucuronidase. CD anal. (in the 200-250-nm range) of the oligosaccharides showed that the CD spectrum of hyaluronate in aq. soln. at neutral pH does not reflect to any substantial degree a polymer conformation which requires cooperative interaction between several repeating residues for stabilization. The enhanced CD properties of hyaluronate relative to monosaccharides are primarily related to the existence of the .beta.-1,4 linkage from N-acetyl-D-glucosamine to D-glucuronate. Chondroitin, the N-acetyl-D-galactosamine analog of hyaluronate, was prep'd. by chem. desulfation of chondroitin 4- and 6-sulfates. The purified product had a mol. wt. range of 4000-8000 (10-20 disaccharide units). It was digested with testicular hyaluronidase, and the split products were isolated by gel filtration. In contrast to hyaluronate, the cleavage products included both the preponderant analogous repeating disaccharide multiples with N-acetyl-D-galactosamine at the reducing end and smaller quantities of oligosaccharides composed of an odd no. of monosaccharides. These latter products were presumably derived from the ends of the shortened chondroitin chains, resulting from methanolysis during the desulfation and alk. elimination of reducing hexosamines. CD spectroscopic anal. of chondroitin relative to its constituent monosaccharides showed that this glycosaminoglycan does not share the structural feature which results in substantially enhanced CD properties for hyaluronate. A hypothesis relating the CD properties of N-acetylated hexosamines in oligo- and polysaccharides to the dihedral angle about the C-O bond at C3 may explain the CD dependence on both the hexosaminidic linkage and hexosamine configuration at C4.

17 ANSWER 1 OF 5 CAPLUS COPYRIGHT 2003 ACS on STN  
ACCESSION NUMBER: 1999:591819 CAPLUS  
DOCUMENT NUMBER: 131:296939  
TITLE: Purification and characterization of fetal bovine serum .beta.-N-acetyl-D-galactosaminyltransferase and .beta.-D-glucuronyltransferase involved in chondroitin sulfate biosynthesis  
AUTHOR(S): Tsuchida, Kazunori; Lind, Thomas; Kitagawa, Hiroshi; Lindahl, Ulf; Sugahara, Kazuyuki; Lidholt, Kerstin  
CORPORATE SOURCE: Department of Biochemistry, Kobe Pharmaceutical University, Kobe, 658-8558, Japan  
SOURCE: European Journal of Biochemistry (1999), 264(2), 461-467  
CODEN: EJBCAI; ISSN: 0014-2956  
PUBLISHER: Blackwell Science Ltd.  
DOCUMENT TYPE: Journal  
LANGUAGE: English  
AB Chondroitin .beta.-acetylgalactosaminyltransferase II (I) and chondroitin 6-sulfate pentasaccharide .beta.-glucuronyltransferase II (II) are involved in chondroitin sulfate biosynthesis, and transfer an N-acetylgalactosamine (GalNAc) or glucuronic acid (GlcA) residue, resp., through .beta.-linkages to an acceptor chondroitin oligosaccharide derived from the repeating disaccharide region of chondroitin sulfate. They were copurified from fetal bovine serum approx. 2500-fold and 850-fold, resp., by sequential chromatogs. on Red A-agarose, phenyl-Sepharose, S-Sepharose, and wheat germ agglutinin-agarose. Identical and inseparable chromatog. profiles of both glycosyltransferase activities obtained through the above chromatog. steps and gel filtration suggested that the purified enzyme activities are tightly coupled, which could imply a single enzyme with dual transferase activities: I and II, reminiscent of the heparan sulfate polymerase reaction. However, when a polymn. reaction was performed in vitro with the purified serum enzyme prepns. under the polymn. conditions recently developed for the chondroitin-synthesizing system, derived from human melanoma cells, each monosaccharide transfer took place, but no polymn. occurred. These results may suggest that the purified serum enzyme prepns. contains both I and II activities on a single polypeptide or on the resp. polypeptides forming an enzyme complex, but is different from that obtained from melanoma cells in that it transfers a single GalNAc or GlcA residue but does not polymerize chondroitin.  
REFERENCE COUNT: 34 THERE ARE 34 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L17 ANSWER 2 OF 5 CAPLUS COPYRIGHT 2003 ACS on STN  
ACCESSION NUMBER: 1985:200042 CAPLUS  
DOCUMENT NUMBER: 102:200042  
TITLE: Two N-acetylgalactosaminyltransferases are involved in the biosynthesis of chondroitin sulfate  
AUTHOR(S): Rohrmann, Kerstin; Niemann, Reinhard; Buddecke, Eckhart  
CORPORATE SOURCE: Inst. Physiol. Chem., Univ. Muenster, Muenster, D-4400, Fed. Rep. Ger.  
SOURCE: European Journal of Biochemistry (1985), 148(3), 463-9  
CODEN: EJBCAI; ISSN: 0014-2956  
DOCUMENT TYPE: Journal  
LANGUAGE: English  
AB N-Acetylgalactosaminyltransferases I and II (I and II, resp.) were purified from the microsomal fraction of calf arterial tissue and sepd. on Bio-Gel A. I was purified 450-fold. I requires Mn<sup>2+</sup> for maximal activity and transferred N-acetylgalactosamine residues from UDP-[1-3H]GalNAc in .beta.-glycosidic configuration to the nonreducing terminus of the acceptor substrates, GlcA(.beta.1-3)Gal(.beta.1-3)Gal, GlcA(.beta.1-3)Gal(.beta.1-4)Glc, and GlcA(.beta.1-3)Gal. Even-numbered chondroitin oligosaccharides served as acceptors for II,

which transferred N-acetylgalactosamine from UDP-[1-3H]GalNAc to the nonreducing glucuronic acid residues of oligosaccharide acceptor substrates. Max. transfer rates were obtained with a decasaccharide derived from chondroitin. Longer or shorter-chain chondroitin oligosaccharides were less effective acceptor substrates. All reaction products formed by I and II were substrates of .beta.-N-acetylhexosaminidase, which split off the transferred [1-3H]GalNAc completely. In the microsomal fraction, II had a 300-fold higher specific activity than I. In contrast to I, II lost much of its activity during purifn., and underwent rapid thermal denaturation. GlcA-Gal-Gal was a characteristic sequence of the carbohydrate-protein linkage region of proteochondroitin sulfate. The acceptor capacity of this trisaccharide suggested that I is involved in the synthesis of the carbohydrate-protein linkage region. Since II is highly specific for chondroitin oligosaccharides, it apparently participates in chain elongation during chondroitin sulfate synthesis.

L17 ANSWER 3 OF 5 CAPLUS COPYRIGHT 2003 ACS on STN

ACCESSION NUMBER: 1981:116374 CAPLUS

DOCUMENT NUMBER: 94:116374

TITLE: Preparation and circular dichroism analysis of sodium hyaluronate oligosaccharides and chondroitin

AUTHOR(S): Cowman, Mary K.; Balazs, Endre A.; Bergmann, Carl W.; Meyer, Karl

CORPORATE SOURCE: Coll. Physicians Surg., Columbia Univ., New York, NY, 10032, USA

SOURCE: Biochemistry (1981), 20(5), 1379-85

CODEN: BICHAW; ISSN: 0006-2960

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Na hyaluronate was cleaved into a homologous series of oligosaccharides by the action of bovine testicular hyaluronidase (EC 3.2.1.35), an endo-.beta.-hexosaminidase. Digestion conditions and gel-filtration chromatog. fractionation were optimized to produce oligosaccharides, clearly separable into peaks corresponding to 1-23 disaccharide units of the type D-glucuronosyl-N-acetyl-D-glucosamine. The chromatog. method was also employed for the purifn. of a 2nd homologous group of oligosaccharides, with the reversed sequence of monosaccharide units, produced by the action of leech hyaluronidase (EC 3.2.1.36), an endo-.beta.-glucuronidase. CD anal. (in the 200-250-nm range) of the oligosaccharides showed that the CD spectrum of hyaluronate in aq. soln. at neutral pH does not reflect to any substantial degree a polymer conformation which requires cooperative interaction between several repeating residues for stabilization. The enhanced CD properties of hyaluronate relative to monosaccharides are primarily related to the existence of the .beta.-1,4 linkage from N-acetyl-D-glucosamine to D-glucuronate. Chondroitin, the N-acetyl-D-galactosamine analog of hyaluronate, was prep'd. by chem. desulfation of chondroitin 4- and 6-sulfates. The purified product had a mol. wt. range of 4000-8000 (10-20 disaccharide units). It was digested with testicular hyaluronidase, and the split products were isolated by gel filtration. In contrast to hyaluronate, the cleavage products included both the preponderant analogous repeating disaccharide multiples with N-acetyl-D-galactosamine at the reducing end and smaller quantities of oligosaccharides composed of an odd no. of monosaccharides. These latter products were presumably derived from the ends of the shortened chondroitin chains, resulting from methanolysis during the desulfation and alk. elimination of reducing hexosamines. CD spectroscopic anal. of chondroitin relative to its constituent monosaccharides showed that this glycosaminoglycan does not share the structural feature which results in substantially enhanced CD properties for hyaluronate. A hypothesis relating the CD properties of N-acetylated hexosamines in oligo- and polysaccharides to the dihedral angle about the C-O bond at C3 may explain the CD dependence on both the hexosaminidic linkage and hexosamine

configuration at C4.

L17 ANSWER 4 OF 5 MEDLINE on STN  
ACCESSION NUMBER: 1999421651 MEDLINE  
DOCUMENT NUMBER: 99421651 PubMed ID: 10491092  
TITLE: Purification and characterization of fetal bovine serum beta-N-acetyl-D-galactosaminyltransferase and beta-D-glucuronyltransferase involved in chondroitin sulfate biosynthesis.  
AUTHOR: Tsuchida K; Lind T; Kitagawa H; Lindahl U; Sugahara K; Lidholt K  
CORPORATE SOURCE: Department of Biochemistry, Kobe Pharmaceutical University, Japan.  
SOURCE: EUROPEAN JOURNAL OF BIOCHEMISTRY, (1999 Sep) 264 (2) 461-7.  
Journal code: 0107600. ISSN: 0014-2956.  
PUB. COUNTRY: GERMANY: Germany, Federal Republic of  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 199910  
ENTRY DATE: Entered STN: 19991026  
Last Updated on STN: 19991026  
Entered Medline: 19991014  
AB beta-N-Acetylgalactosaminyltransferase II and beta-glucuronyltransferase II, involved in chondroitin sulfate biosynthesis, transfer an N-acetylgalactosamine (GalNAc) and glucuronic acid (GlcA) residue, respectively, through beta-linkages to an acceptor chondroitin oligosaccharide derived from the repeating disaccharide region of chondroitin sulfate. They were copurified from fetal bovine serum approximately 2500-fold and 850-fold, respectively, by sequential chromatographies on Red A-agarose, phenyl-Sepharose, S-Sepharose and wheat germ agglutinin-agarose. Identical and inseparable chromatographic profiles of both glycosyltransferase activities obtained through the above chromatographic steps and gel filtration suggest that the purified enzyme activities are tightly coupled, which could imply a single enzyme with dual transferase activities; beta-N-acetylgalactosaminyltransferase and beta-glucuronyltransferase, reminiscent of the heparan sulfate polymerase reaction. However, when a polymerization reaction was performed in vitro with the purified serum enzyme preparation under the polymerization conditions recently developed for the chondroitin-synthesizing system, derived from human melanoma cells, each monosaccharide transfer took place, but no polymerization occurred. These results may suggest that the purified serum enzyme preparation contains both beta-N-acetylgalactosaminyltransferase II and beta-glucuronyltransferase II activities on a single polypeptide or on the respective polypeptides forming an enzyme complex, but is different from that obtained from melanoma cells in that it transfers a single GalNAc or GlcA residue but does not polymerize chondroitin.

L17 ANSWER 5 OF 5 MEDLINE on STN  
ACCESSION NUMBER: 85203849 MEDLINE  
DOCUMENT NUMBER: 85203849 PubMed ID: 3922754  
TITLE: Two N-acetylgalactosaminyltransferase are involved in the biosynthesis of chondroitin sulfate.  
AUTHOR: Rohrmann K; Niemann R; Buddecke E  
SOURCE: EUROPEAN JOURNAL OF BIOCHEMISTRY, (1985 May 2) 148 (3) 463-9.  
Journal code: 0107600. ISSN: 0014-2956.  
PUB. COUNTRY: GERMANY, WEST: Germany, Federal Republic of  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 198507  
ENTRY DATE: Entered STN: 19900320

Last Updated on STN: 19900320  
Entered Medline: 19850705

AB Two N-acetylgalactosaminyltransferases, designated I and II, have been purified from the microsomal fraction of calf arterial tissue and separated on Bio-Gel A. N-Acetylgalactosaminyltransferase I was purified 450-fold. It requires Mn<sup>2+</sup> for maximal activity and transfers N-acetylgalactosamine residues from UDP-[1-3H]GalNAc in beta-glycosidic configuration to the non-reducing terminus of the acceptor substrates GlcA(beta 1-3)Gal(beta 1-3)Gal, GlcA(beta 1-3)Gal(beta 1-4)Glc and GlcA(beta 1-3)Gal. Even-numbered chondroitin oligosaccharides serve as acceptors for N-acetylgalactosaminyltransferase II, which transfers N-acetylgalactosamine from UDP-[1-3H]GalNAc to the non-reducing glucuronic acid residues of oligosaccharide acceptor substrates. Maximum transfer rates were obtained with a decasaccharide derived from chondroitin. Longer or shorter-chain chondroitin oligosaccharides are less effective acceptor substrates. All reaction products formed by N-acetylgalactosaminyltransferases I and II are substrates of beta-N-acetylhexosaminidase, which splits off the transferred [1-3H]GalNAc completely. In the microsomal fraction N-acetylgalactosaminyltransferase II had a 300-fold higher specific activity than N-acetylgalactosaminyltransferase I. In contrast to enzyme I, enzyme II loses much of its activity during the purification procedure and undergoes rapid thermodenaturation. GlcA-Gal-Gal is a characteristic sequence of the carbohydrate-protein linkage region of proteochondroitin sulfate. The acceptor capacity of this trisaccharide suggests that N-acetylgalactosaminyltransferase I is involved in the synthesis of the carbohydrate-protein linkage region. Since N-acetylgalactosaminyltransferase II is highly specific for chondroitin oligosaccharides, we conclude that it participates in chain elongation during chondroitin sulfate synthesis.

d his

(FILE 'HOME' ENTERED AT 15:11:28 ON 03 SEP 2003)

FILE 'CAPLUS, MEDLINE' ENTERED AT 15:11:36 ON 03 SEP 2003

L1           0 S CHRONDROITIN POLYMER  
L2           10 S CHONDROITIN POLYMER  
L3           0 S L2 AND COMPOSITION  
L4           7 S L2 AND CHONDROITIN SYNTHASE  
L5           3 S L2 AND SULFATED  
L6           2 DUP REM L5 (1 DUPLICATE REMOVED)  
L7           7 S L2 NOT L5  
L8           8 S CHONDROITIN POLYSACCHARIDE  
L9           6 DUP REM L8 (2 DUPLICATES REMOVED)  
L10          23094 S CHONDROITIN  
L11          35 S L10 AND CHONDROITIN SYNTHASE  
L12          11 S L11 AND RECOMBINANT  
L13          24 S L11 NOT L12  
L14          10 S L13 AND PASTEURELLA  
L15          20 S CHONDROITIN OLIGOSACCHARIDE  
L16          0 S L15 AND PURIFIED COMPOSITION  
L17          5 S L15 AND PURIFIED

L9 ANSWER 1 OF 5 CAPLUS COPYRIGHT 2003 ACS on STN  
 ACCESSION NUMBER: 2002:487738 CAPLUS  
 DOCUMENT NUMBER: 137:59509  
 TITLE: DNA and protein sequences of UDP-N-acetylglucosamine  
       4-epimerase from *Bacillus* and *Neisseria* and their uses  
 INVENTOR(S): Endo, Tetsuo; Koizumi, Satoshi; Tabata, Kazuhiko;  
               Ozaki, Akio  
 PATENT ASSIGNEE(S): Kyowa Hakko Kogyo Co., Ltd., Japan  
 SOURCE: PCT Int. Appl., 59 pp.  
 CODEN: PIXXD2  
 DOCUMENT TYPE: Patent  
 LANGUAGE: Japanese  
 FAMILY ACC. NUM. COUNT: 1  
 PATENT INFORMATION:

| PATENT NO.             | KIND                                                                                                                                                                                                                                                                                                                                                                                                              | DATE     | APPLICATION NO. | DATE       |
|------------------------|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|----------|-----------------|------------|
| WO 2002050267          | A1                                                                                                                                                                                                                                                                                                                                                                                                                | 20020627 | WO 2001-JP11269 | 20011221   |
| W:                     | AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN,<br>CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH,<br>GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KR, KZ, LC, LK, LR, LS,<br>LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL,<br>PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ, UA,<br>UG, US, UZ, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM |          |                 |            |
| RW:                    | GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, CH,<br>CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR,<br>BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG                                                                                                                                                                                                              |          |                 |            |
| AU 2002025352          | A5                                                                                                                                                                                                                                                                                                                                                                                                                | 20020701 | AU 2002-25352   | 20011221   |
| PRIORITY APPLN. INFO.: |                                                                                                                                                                                                                                                                                                                                                                                                                   |          | JP 2000-388992  | A 20001221 |
|                        |                                                                                                                                                                                                                                                                                                                                                                                                                   |          | WO 2001-JP11269 | W 20011221 |

AB This invention provides the process of prepn. of UDP-N-acetylglucosamine  
       4-epimerase from *Bacillus* and *Neisseria*. The DNA and protein sequences of  
       UDP-N-acetylglucosamine 4-epimerase from *Bacillus subtilis* and *Neisseria*  
       gonorrhoeae were disclosed. The invention provides detailed description  
       about plasmid construction for UDP-N-acetylglucosamine 4-epimerase  
       expression in *E. coli*. The UDP-N-acetylglucosamine 4-epimerase can be  
       used for biosynthesis of UDP-N-acetylgalactosamine and saccharide contg.  
       N-acetylgalactosamine.

REFERENCE COUNT: 11 THERE ARE 11 CITED REFERENCES AVAILABLE FOR THIS  
                   RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L9 ANSWER 2 OF 5 CAPLUS COPYRIGHT 2003 ACS on STN  
 ACCESSION NUMBER: 2001:195197 CAPLUS  
 DOCUMENT NUMBER: 134:227437  
 TITLE: Biocompatible surfaces comprising  
       polysaccharide derivatives and a method for  
       their preparation  
 INVENTOR(S): Nelson, Deanna J.; Hai, Ton That; Pereira, David E.;  
               Estep, Timothy N.  
 PATENT ASSIGNEE(S): Baxter International Inc., USA  
 SOURCE: U.S., 19 pp.  
 CODEN: USXXAM  
 DOCUMENT TYPE: Patent  
 LANGUAGE: English  
 FAMILY ACC. NUM. COUNT: 1  
 PATENT INFORMATION:

| PATENT NO.             | KIND | DATE     | APPLICATION NO. | DATE     |
|------------------------|------|----------|-----------------|----------|
| US 6204254             | B1   | 20010320 | US 1997-928841  | 19970912 |
| PRIORITY APPLN. INFO.: |      |          | US 1997-928841  | 19970912 |

AB A novel group of compds. is disclosed for decorating the surface of  
       synthetic polymeric or tissue derived prostheses to prevent adverse

rejection events. The decorating mols. are obtained as derivs. of naturally occurring polysaccharides, derivatized to provide functionally reactive groups at the termini thereof, and the reacting with nucleophilic or other groups on the surface of the prosthesis in a simple one step reaction. Some of these reagents are useful in noncovalent adsorption to polyolefinic or perfluorocarbon based materials. Finally, phospholipids partially substituted with the nonantigenic polysaccharides provide a superior bipolar component for liposome formation. Chondroitin sulfate-modified-distearoyl phosphatidylethanolamine (I) was prep'd. by reaction of chondroitin sulfate-CO-N-oxysuccinimide with lyso-distearoyl phosphatidylethanolamine. Liposomes were prep'd. by micro-fluidization (emulsification) of a compn. of I/hydrogenated soy phosphatidylcholine/cholesterol in molar proportions of 5:55:40, resp. It is anticipated that the blood circulation half-lives of the biocompatible liposomes will be significantly longer than those of liposomes formulated without the I.

REFERENCE COUNT: 11 THERE ARE 11 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L9 ANSWER 3 OF 5 CAPLUS COPYRIGHT 2003 ACS on STN  
 ACCESSION NUMBER: 1999:77464 CAPLUS  
 DOCUMENT NUMBER: 130:158447  
 TITLE: Therapeutic hemoglobin-polysaccharide complexes having isotropically increased size and masked antigenicity  
 INVENTOR(S): Hai, Ton That; Pereira, David E.; Nelson, Deanna J.  
 PATENT ASSIGNEE(S): Baxter International Inc., USA  
 SOURCE: PCT Int. Appl., 51 pp.  
 CODEN: PIXXD2  
 DOCUMENT TYPE: Patent  
 LANGUAGE: English  
 FAMILY ACC. NUM. COUNT: 1  
 PATENT INFORMATION:

| PATENT NO.                                                                 | KIND | DATE     | APPLICATION NO.   | DATE     |
|----------------------------------------------------------------------------|------|----------|-------------------|----------|
| WO 9903484                                                                 | A1   | 19990128 | WO 1998-US12941   | 19980622 |
| W: AU, CA, JP                                                              |      |          |                   |          |
| RW: AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE |      |          |                   |          |
| US 5981710                                                                 | A    | 19991109 | US 1997-896743    | 19970721 |
| AU 9881586                                                                 | A1   | 19990210 | AU 1998-81586     | 19980622 |
| AU 742849                                                                  | B2   | 20020117 |                   |          |
| EP 1017405                                                                 | A1   | 20000712 | EP 1998-931462    | 19980622 |
| R: DE, FR, GB                                                              |      |          |                   |          |
| JP 2001510202                                                              | T2   | 20010731 | JP 2000-502782    | 19980622 |
| PRIORITY APPLN. INFO.:                                                     |      |          | US 1997-896743 A  | 19970721 |
|                                                                            |      |          | WO 1998-US12941 W | 19980622 |

AB Novel polysaccharide compds. are disclosed for decorating biomol. surfaces to increase isotropic size and mask antigenicity. The oligosaccharides may be synthesized as repeating disaccharide units, or may be derived by acid hydrolysis of naturally occurring polysaccharides. Such natural sources include chondroitins obtained from shark cartilage, or hyaluronic acid. The polyanionic sulfate groups contained in the sugar moieties impart neg. charges which repel the mols. from the neg. charged wall of capillaries, to lengthen retention times of decorated drug mols., such as cross-linked Hb, in the peripheral circulation.

REFERENCE COUNT: 3 THERE ARE 3 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L9 ANSWER 4 OF 5 CAPLUS COPYRIGHT 2003 ACS on STN  
 ACCESSION NUMBER: 1996:72306 CAPLUS  
 DOCUMENT NUMBER: 124:110772

TITLE: Biosynthesis of dermatan sulfate. Defructosylated Escherichia coli K4 capsular polysaccharide as a substrate for the D-glucuronyl C-5 epimerase, and an indication of a two-base reaction mechanism

AUTHOR(S): Hannesson, Helgi H.; Hagner-McWhirter, Asa; Tiedemann, Kerstin; Lindahl, Ulf; Malmstroem, Anders

CORPORATE SOURCE: Dep. Med. Physiological Chemistry, Univ. Uppsala, Uppsala, S-751 23, Swed.

SOURCE: Biochemical Journal (1996), 313(2), 589-96

CODEN: BIJOAK; ISSN: 0264-6021

PUBLISHER: Portland Press

DOCUMENT TYPE: Journal

LANGUAGE: English

AB The capsular polysaccharide from Escherichia coli K4 consists of a chondroitin  $\{[\text{GlcA}(\beta.1.\text{fwdarw.3})\text{GalNAc}(\beta.1.\text{fwdarw.4})]_n\}$  backbone, to which  $\beta$ -fructofuranose units are linked to C-3 of D-glucuronic acid (GlcA) residues. Removal of the fructose units by mild acid hydrolysis provided a substrate for the GlcA C-5 epimerase, which is involved in the generation of L-iduronic acid (IdoA) units during dermatan sulfate biosynthesis. Incubation of this substrate with solubilized fibroblast microsomal enzyme in the presence of  $^3\text{H}_2\text{O}$  resulted in the incorporation of tritium at C-5 of hexuronyl units. A  $K_m$  of  $67 \times 10^{-6}$  M hexuronic acid (equiv. to disaccharide units) was detd., which is similar to that ( $80 \times 10^{-6}$  M) obtained for dermatan (desulfated dermatan sulfate).  $V_{max}$  was about 4 times higher with dermatan than with the K4 substrate. A defructosylated K4 polysaccharide isolated after incubation of bacteria with D-[5- $^3\text{H}$ ] glucose released  $^3\text{H}_2\text{O}$  on reaction with the epimerase, and thus could be used to assay the enzyme. Incubation of a K4 substrate with solubilized microsomal epimerase for 6 h in the presence of  $^3\text{H}_2\text{O}$  resulted in the formation of about 5% IdoA and approx. equal amts. of  $^3\text{H}$  in GlcA and IdoA. A corresponding incubation of dermatan yielded approx. 22% GlcA, which contained virtually all the  $^3\text{H}$  label. These results are tentatively explained in terms of a two-base reaction mechanism, involving a monoprotic L-ido-specific base and a polyprotic D-gluco-specific base. Most of the IdoA residues generated by the enzyme occurred singly, although some formation of two or three consecutive IdoA-contg. disaccharide units was obsd.

L9 ANSWER 5 OF 5 MEDLINE on STN

ACCESSION NUMBER: 96152546 MEDLINE

DOCUMENT NUMBER: 96152546 PubMed ID: 8573097

TITLE: Biosynthesis of dermatan sulphate. Defructosylated Escherichia coli K4 capsular polysaccharide as a substrate for the D-glucuronyl C-5 epimerase, and an indication of a two-base reaction mechanism.

AUTHOR: Hannesson H H; Hagner-McWhirter A; Tiedemann K; Lindahl U; Malmstrom A

CORPORATE SOURCE: Department of Medical and Physiological Chemistry, University of Uppsala, Sweden.

SOURCE: BIOCHEMICAL JOURNAL, (1996 Jan 15) 313 ( Pt 2) 589-96.  
Journal code: 2984726R. ISSN: 0264-6021.

PUB. COUNTRY: ENGLAND: United Kingdom

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 199603

ENTRY DATE: Entered STN: 19960315  
Last Updated on STN: 19980206  
Entered Medline: 19960301

AB The capsular polysaccharide from Escherichia coli K4 consists of a chondroitin  $\{[\text{GlcA}(\beta 1\rightarrow 3)\text{GalNAc}(\beta 1\rightarrow 4)]_n\}$  backbone, to which beta-fructofuranose units are linked to C-3 of D-glucuronic acid (GlcA) residues. Removal of the fructose units by mild acid hydrolysis

provided a substrate for the GlcA C-5 epimerase, which is involved in the generation of L-iduronic acid (IdoA) units during dermatan sulphate biosynthesis. Incubation of this substrate with solubilized fibroblast microsomal enzyme in the presence of  $^3\text{H}_2\text{O}$  resulted in the incorporation of tritium at C-5 of hexuronyl units. A  $K_m$  of  $67 \times 10^{-6}$  M hexuronic acid (equivalent to disaccharide units) was determined, which is similar to that ( $80 \times 10^{-6}$  M) obtained for dermatan (desulphated dermatan sulphate).  $V_{max}$  was about 4 times higher with dermatan than with the K4 substrate. A defructosylated K4 polysaccharide isolated after incubation of bacteria with D-[5- $^3\text{H}$ ] glucose released  $^3\text{H}_2\text{O}$  on reaction with the epimerase, and thus could be used to assay the enzyme. Incubation of a K4 substrate with solubilized microsomal epimerase for 6 h in the presence of  $^3\text{H}_2\text{O}$  resulted in the formation of about 5% IdoA and approximately equal amounts of  $^3\text{H}$  in GlcA and IdoA. A corresponding incubation of dermatan yielded approx. 22% GlcA, which contained virtually all the  $^3\text{H}$  label. These results are tentatively explained in terms of a two-base reaction mechanism, involving a monoprotic L-ido-specific base and a polyprotic D-gluco-specific base. Most of the IdoA residues generated by the enzyme occurred singly, although some formation of two or three consecutive IdoA-containing disaccharide units was observed.

d his

(FILE 'HOME' ENTERED AT 17:40:31 ON 04 SEP 2003)

FILE 'CAPLUS, MEDLINE' ENTERED AT 17:40:41 ON 04 SEP 2003

L1           0 S CHONDROITIN EPIMER  
L2           1 S EPIMERIZATION OF CHONDROITIN  
L3           23106 S CHONDROITIN  
L4           1 S L3 AND EPIMERS  
L5           1158 S IDURONIC ACID  
L6           104 S L5 AND GLUCOSE  
L7           21 S L6 AND POLYMER  
L8           31 S L6 AND POLYSACCHARIDE  
L9           5 S L8 AND CHONDROITIN